

A community-level test of the leaf-height-seed ecology strategy scheme in relation to grazing conditions

Golodets, Carly*¹; Sternberg, Marcelo^{2,3} & Kigel, Jaime^{1,4}

¹Robert H. Smith Institute for Plant Sciences and Genetics in Agriculture, Faculty of Agricultural, Food and Environmental Quality Sciences, The Hebrew University of Jerusalem, PO Box 12, 76100 Rehovot, Israel;

²Department of Plant Sciences, Faculty of Life Sciences, Tel Aviv University, 69978 Tel Aviv, Israel; ³Email marcelos@tauex.tau.ac.il; ⁴Email kigel@agri.huji.ac.il;

*Corresponding author; Fax +972 8 9467763; Email adonis@agri.huji.ac.il

Abstract

Question: Is the assumption of trait independence implied in Westoby's (1998) leaf-height-seed (LHS) ecology strategy scheme upheld in a Mediterranean grazing system dominated by annuals? Is the LHS approach applicable at the community level?

Location: Northern Israel.

Methods: LHS traits (specific leaf area [SLA], plant height and seed mass), and additional leaf traits (leaf dry matter content [LDMC], leaf area, and leaf content of nitrogen [LNC], carbon [LCC], and phosphorus [LPC]), were analyzed at the species and community levels. Treatments included manipulations of grazing intensity (moderate and heavy) and protection from grazing. We focused on species comprising 80% of biomass over all treatments, assuming that these species drive trait relationships and ecosystem processes.

Results: At the species level, SLA and seed mass were negatively correlated, and plant height was positively correlated to LCC. SLA, seed mass, and LPC increased with protection from grazing. At the community level, redundancy analysis revealed one principal gradient of variation: SLA, correlated to grazing, versus seed mass and plant height, associated with protection from grazing. We divided community functional parameters into two groups according to grazing response: (1) plant height, seed mass, LDMC, and LCC, associated with protection from grazing, and (2) SLA, associated with grazing.

Conclusions: The assumption of independence between LHS traits was not upheld at the species level in this Mediterranean grazing system. At the community level, the LHS approach captured most of the variation associated with protection from grazing, reflecting changes in dominance within the plant community.

Keywords: Community functional parameters; Mediterranean grassland; Plant traits; Redundancy analysis.

Nomenclature: Feinbrun-Dothan & Danin (1991).

Abbreviations: LHS = Leaf-Height-Seed, SLA = Specific leaf area, LDMC = Leaf dry matter content, RDA = Redundancy analysis, LNC = Leaf nitrogen content, LCC = Leaf carbon content, LPC = Leaf phosphorus content.

Introduction

Plant ecology strategy schemes enable the categorization of plant species responses to disturbance, competitive ability, and dispersal capability, and their roles in ecosystem functioning. This categorization facilitates reduction of the diversity of plant species responses to a few, easily defined categories with meaningful ecological significance. Two major dimensions of ecological variation are the ability to cope with disturbance and adaptation to fast versus slow growth opportunities (Westoby et al. 2002). The current challenge is to define how these dimensions are represented in plant response traits, and to link them to ecosystem functioning (Weiher et al. 1999). Raunkiaer's (1937) life form scheme has been widely used for decades and its life form definitions are incorporated into vegetation analyses as a matter of course. Grime's (1974, 2001) competitive-stress tolerant-ruderal (C-S-R) scheme is also frequently used. More recently, Westoby (1998) proposed the leaf-height-seed (LHS) scheme as an alternative method to summarize all major dimensions of variation in plant responses to their environment. This scheme was developed for use at global scales, and is therefore based on easily measured plant traits: canopy height, specific leaf area, and seed mass. Each of these three traits represents a fundamental trade-off that controls plant strategies, so that differences in trait values between species are

assumed to be ecologically meaningful. Weiher et al. (1999) concluded that the best traits for describing plant fitness are relative growth rate (RGR), seed mass, and plant height. This corresponds well to Westoby's (1998) LHS strategy scheme, since SLA is a good surrogate for potential RGR (Westoby et al. 1996; Westoby 1998; Wright & Westoby 1999).

The LHS scheme has been applied to various situations, such as plant communities among different habitats (Lavergne et al. 2003), a successional gradient (Garnier et al. 2004), and a diversity of management strategies (Moog et al. 2005). In these studies, the scheme captured the main gradients of variation in the vegetation. However, seed mass and canopy height were strongly correlated, thus contradicting the assumption of independence between the three dimensions of the model (Westoby 1998, 1999). Furthermore, the scheme did not adequately discriminate changes in vegetation composition (Lavergne et al. 2003; Moog et al. 2005). A common conclusion was that additional dimensions of variation are necessary, especially for use at a global scale (Garnier et al. 2004; Diaz et al. 2006).

We tested the LHS scheme to assess the impact of changing land use in a Mediterranean grassland dominated by annual species, using an approach fundamentally different from that of Lavergne et al. (2003) and Garnier et al. (2004). These authors divided species into well-defined categories (e.g., successional stages), and did not consider overlaps in species composition. In their systems, comparisons can be species-based, since each species belongs to a particular plant community. In our system, there is significant overlap in species composition and frequency between the communities, therefore we adopted a continuous approach (Moog et al. 2005), focusing on differences between communities based on species frequency, rather than differences in species identity. As proposed by Garnier et al. (2007), we used community functional parameters (*sensu* Violle et al. 2007), which are aggregated trait values of the species constituting most of the community biomass (>80%), weighted according to their relative biomass. This approach follows the "biomass ratio hypothesis" of Grime (1998), which proposes that ecosystem functioning is primarily determined by the species that dominate plant biomass production.

We studied the applicability of the LHS scheme in a Mediterranean cattle-grazing system, using redundancy analysis to examine variation in community functional parameters, and to determine the effects of grazing intensity. Westoby (1999) offered predictions about grazing effects on LHS traits in a

grassland system with non-selective livestock grazing: (a) dominance by high-SLA species that exhibit fast re-growth and leaf turnover, with a shift to low-SLA species upon release from grazing pressure; (b) heavy grazing favors shorter plants over taller ones in dense grasslands, while release from grazing favors taller plants due to increased competition for light; and (c) grazing should promote species that produce large quantities of small seeds to maximize opportunities for seedling establishment in the disturbed environment, while release from grazing should promote a shift towards species with larger seeds, because larger seed size confers an advantage for germination under suboptimal conditions. We tested these three predictions by examining the responses of LHS traits to protection from grazing. We also investigated whether the assumptions of the LHS strategy scheme hold at the species level in our grazing system, and then examined the scheme at the community level using community functional parameters. In addition, we examined the variation and relevance of other functional parameters, based on morphological and chemical leaf traits, to develop a more inclusive perspective on functional relationships within this grassland community.

We asked the following questions:

- (1) Do the three traits of the LHS strategy scheme adequately represent variations in species composition caused by changes in grazing conditions?
- (2) Does the community-level approach reflect changes in dominance in this Mediterranean grassland community due to protection from grazing? In addition, does the predominance of grasses drive the community-level response?
- (3) Do additional quantitative leaf traits improve the ability of the model to explain the changes in community composition due to protection from grazing?

Materials and Methods

Site description

The research was carried out at the Karei Deshe Experimental Station (32°55'N, 35°35'E, 150 m a.s.l., 570 mm annual rainfall) in the northeast Galilee region of Israel. The vegetation is classified as Mediterranean semi-steppe batha (Zohary 1973), dominated by grasses and forbs. The dominant perennial species are the hemicryptophytes *Bituminaria bituminosa*, *Echinops gaillardotii*, *E. adenocaulos*,

Ferula communis, and *Hordeum bulbosum*, forming 40-60% of the cover (Noy-Meir et al. 1989; Sternberg et al. 2000). Most other species are annuals, including grasses (*Avena sterilis*, *Alopecurus utriculatus*, *Bromus* spp.), legumes (*Medicago* spp., *Trifolium* spp.), and several composites, crucifers, and umbellifers. Growth and development of the vegetation depends almost entirely on seasonal rainfall, from mid-October/late November to late April/early May. Productivity is strongly dependent on the amount and distribution of this rainfall.

Experimental treatments

The rangeland at the station is grazed by cattle, under a controlled grazing system (Sternberg et al. 2000). We selected two grazing treatments – continuous heavy (CH), and continuous moderate (CM), with 1.1 and 0.55 cows · ha⁻¹ · year⁻¹, respectively, and plots protected from grazing for 30-40 years (long-term protection, LP). The cows grazed for approximately 190 days per year of the research, from mid-January to late August. Each experimental treatment included two replicate plots, giving a total of six plots for the experiment. The grazed plots are relatively large compared to the protected plots (ca. 20-30 ha versus 0.4-2.0 ha). The actual sampled area, however, was very similar between treatments and shared similar environmental characteristics (slope, rockiness). Within each grazed plot, five 10 m × 10 m exclosures (i.e., five replicates per plot) were established in March 2003, separated from each other by 50-100 m. In the smaller, protected plots, five 2.5-m stakes were placed randomly within each plot, marking the center of the 100 m² sampling area.

Sampling

Vegetation was sampled in the spring (April) of 2003 and 2004, from five 25 cm × 25 cm quadrats within each exclosure in the grazed plots and within a ca. 6 m radius of each stake in the protected plots. Herbaceous vegetation was sampled randomly within the sampling area, avoiding rocks and large, relatively dispersed perennial hemicryptophytes (i.e., *Echinops* spp., *F. communis*, *B. bituminosa*), but including the ubiquitous perennial grass *Hordeum bulbosum*. Vegetation sampling involved complete removal of all aboveground plant material within the sampling quadrat. Plants were sorted to species level, identified, counted, and dried at 70°C for 48 h.

Plant traits were sampled for the most abundant species that cumulatively (in decreasing abundance) comprised at least 80% of the biomass in each plot in the treatments where they occurred. Pakeman & Queded (2007) have shown that this approach can provide adequate estimates of community functional parameters for quantitative traits if trait values do not vary greatly between species. Lists of species satisfying the 80% biomass requirement in each year of the research, and of biomass percentages, are presented in Table S1 and Table S2, respectively (see Supporting Information section for details of Tables S1 and S2). Biomass values ranged from ~300 g m⁻² in grazed plots to ~700 g m⁻² in ungrazed plots.

Traits measured included: (1) LHS traits, namely, specific leaf area (SLA mm² g⁻¹), maximum canopy height (cm), and seed mass (mg seed⁻¹), and (2) other selected leaf traits, namely, leaf dry matter content (LDMC mg g⁻¹), leaf area (mm²), leaf nitrogen content (LNC mg g⁻¹), leaf carbon content (LCC mg g⁻¹), and leaf phosphorus content (LPC mg g⁻¹), the latter three being summarized as “leaf chemical traits.” All morphological traits were sampled once in 2003 or 2004. Leaf chemical traits were sampled during 2003. For all trait measurements, excluding seed data, replication was at the level of the plant (within each species). Methods for determination of leaf traits followed Cornelissen et al. (2003). Petioles were omitted for leaf trait determination to enable comparisons between species with petiolate and sessile leaves. Plant height was measured at peak biomass as the maximum height of foliage on mature, reproductive plants, including leaves on inflorescences and scapes. Heights for the late-flowering *Scolymus maculatus* and *Carthamus glaucus* were measured in early summer (late May). Leaf morphological traits and plant heights were measured on three to five plants per species per plot per treatment (six to ten replicates (exclosures) per treatment); leaf chemical traits were measured on one to two plants per species per plot per treatment (two to four replicates (exclosures) per treatment). Seed mass was determined on an analytical balance (± 0.1 mg) by weighing three to five replicates of ten seeds per species per plot per treatment (six to ten replicates per treatment). Each replicate was a pooled sample from up to five plants.

Grazing intensity

We measured grazing intensity by calculating green biomass production in exclosures (immediately

after their construction) as a percentage of green biomass production in long-term protected plots, then the result subtracted from 100%. Grazing intensity was determined as 65% for the heavy treatment, and 31% for the moderate treatment.

Statistical analysis

For redundancy analysis (RDA) and other statistical analyses, community functional parameters were calculated per plot based on the species comprising 80% of the total biomass. For the initial LHS and quantitative trait analyses, species' biomass values were pooled over the 2 years to maximize the number of species for the RDA. An initial, repeated-measures ANOVA on community functional parameters, with time as the repeated measure, did not reveal any effect of year on community functional parameters or on their responses to protection from grazing, so we believe that the decision to pool the data is justified. This pooling produced a dataset of 15 species (2003-2004 dataset; pooled species lists from Table S1). In the leaf chemical analyses, biomass percentages of 11 species from the 2003 dataset (*Lolium rigidum* was excluded due to lack of leaf material) were used. In both dataset analyses, the total biomass of the species concerned was set at 100%, and percentage biomass re-calculated for each species. Traits were defined as stable (SLA, seed mass, LDMC, LCC) or variable (plant height, LNC, LPC, leaf area), *sensu* Garnier et al. (2007), for calculation of community functional parameters. Stable traits were represented by a cross-treatment average value, while for variable traits we used per treatment values.

For both datasets, analysis of variance (ANOVA) was conducted on ln-transformed (SLA, plant height, seed mass, LDMC, leaf area) or arcsine-transformed (LNC, LCC, LPC) trait data to test the effect of protection from grazing. We used a nested, mixed-model ANOVA, with treatment as a fixed effect, while the random effects were plot nested in treatment, and species nested in plot nested in treatment. Since eight of the 15 species were grasses, we tested whether there was a "grass effect" driving responses of community functional parameters to protection from grazing. Community functional parameters (with plots as replicates) were analyzed by analysis of covariance (ANCOVA), with treatment as the main factor and percentage grasses as a covariate; results were compared with those of the ANOVA. Of all the parameters tested (from both datasets), there were no instances where percentage grasses cancelled a significant response to protection

from grazing, therefore, we chose to present only the ANOVA results. Nevertheless, there was one instance where percentage grasses masked a significant grazing response; this is explained in detail in the Results section. For all of the above analyses, Tukey HSD was used for comparison of means when effects were significant. At the species level, Pearson correlations were conducted on pairs of traits, using species average trait values. Data were transformed to natural logarithms (SLA, plant height, seed mass, LDMC, and leaf area) or arcsine values (LNC, LCC, and LPC). At the community level, Spearman correlations (due to different scales) were conducted for pairs of community functional parameters, on a per plot basis. Spearman correlations were also conducted between community functional parameters and grazing intensity. ANOVA, ANCOVA, and correlations were conducted in JMP IN 5.1 (SAS Institute Inc., Cary, NC, USA).

A detrended correspondence analysis (DCA) of species biomass data yielded short axis lengths (2.043 SD for Axis 1, 0.718 SD for Axis 2), indicating that the species gradient sampled would best be analyzed by a linear ordination method (Lepš & Šmilauer 2003). We used redundancy analysis (RDA), a direct (constrained), linear, ordination method that includes explanatory variables for interpreting gradients of variation. RDA ordinations were conducted with CANOCO for Windows 4.5 (Ter Braak & Šmilauer 2002a). Each ordination was based on a "species" matrix, comprising plots ("samples" in CANOCO) by community functional parameters ("species" in CANOCO), and an "environment" matrix, comprising plots by environmental variables (i.e., grazing intensity). The number of community functional parameters in each ordination ranged from three (LHS) to six (LHS and leaf chemical traits). The "plot×community functional parameter" matrix was obtained by multiplying a "plot×species" matrix by a "species×community functional parameter" matrix. We conducted ordinations on untransformed data, which were centered and standardized. We ran a Monte Carlo test (999 permutations) to determine whether grazing intensity explained a significant amount of variation along the first canonical axis. We chose restricted permutations to test for treatment effects, using the split-plot feature. Species biomass values were added as supplementary environmental variables (plot×species matrix) to the RDA of LHS community functional parameters, to aid interpretation. Ordination plots were created in CanoDraw for Windows 4.0 (Ter Braak & Šmilauer 2002b).

Results

Morphological data analysis presented here is based on the combined 2003-2004 dataset, while the analysis of chemical traits and their relationships with morphological traits are based on the 2003 dataset.

LHS trait analysis

The morphological trait analysis included LHS traits (SLA, plant height, and seed mass), LDMC, and leaf area. At the species level, SLA and seed mass were negatively and significantly correlated to each other (Table 1), compared to a marginal positive correlation between plant height and seed mass, and a marginal negative correlation between LDMC and leaf area. The response to protection from grazing (i.e., high (CH) and moderate (CM) grazing intensity versus long-term (LP) protection from grazing) was significant and positive for SLA and seed mass, while LDMC decreased significantly with protection from grazing compared to the moderate grazing treatment (Table 2; Fig. 1).

At the community level, the functional parameters of plant height and seed mass significantly increased with protection from grazing, while the SLA parameter exhibited a non-significant decrease (Table 3; Fig. 1). RDA revealed one principal gradient of variation. SLA was positively associated with grazing intensity and with two annual species: the thistle *Scolymus maculatus* and the crucifer *Ra-*

pistrum rugosum. At the other end of the gradient was the perennial grass, *Hordeum bulbosum*, which was positively associated with the protected plots. At the community level, SLA was strongly and negatively correlated to seed mass, plant height, and LDMC, all of which were positively associated with protected plots and negatively correlated with grazing intensity (Table 4; Fig. 2). Consequently, there were positive significant correlations between plant height, seed mass, and LDMC. The three LHS parameters were significantly correlated to the first RDA axis, while leaf area was marginally correlated to Axis 2 (Table 4).

Morphological and chemical trait analysis

This analysis included LHS traits and leaf chemical traits (LNC, LCC, and LPC) sampled from 11 species in 2003. Protection from grazing increased LPC at the species level (Table 2). At the community level, protection from grazing decreased SLA, and increased plant height and LCC (Table 3). In the analysis of effects of grass abundance on traits, LPC was the only trait showing a significant negative correlation to grass abundance (% grasses: $F_{1,2} = 858$, $P = 0.001$). This correlation masked an underlying grazing response, such that LPC at moderate grazing intensity or under protection from grazing was significantly greater than at high grazing intensity ($F_{2,2} = 333$, $P = 0.003$). At the species level, plant height was positively and significantly correlated to LCC, while the negative correlation to LPC

Table 1. Species-level analysis: Correlation coefficients among plant traits. Morphological traits analyzed for the 2003-2004 dataset (15 species) were specific leaf area (SLA), plant height, seed mass, leaf dry matter content (LDMC), and leaf area. Chemical and morphological traits analyzed for the 2003 dataset (11 species) were SLA, plant height, seed mass, leaf nitrogen content (LNC), leaf carbon content (LCC), and leaf phosphorus content (LPC). Bold values are significant ($\alpha = 0.05$); italicized values are marginally significant ($\alpha = 0.1$).

2003-2004 dataset:	Morphological traits					
	SLA	Plant height	Seed mass	LDMC	Leaf area	
SLA	1					
Plant height	-0.346	1				
Seed mass	-0.572	<i>0.517</i>	1			
LDMC	-0.384	0.028	0.055	1		
Leaf area	-0.408	0.272	0.261	<i>-0.489</i>	1	

2003 dataset:	Morphological and chemical traits					
	SLA	Plant height	Seed mass	LNC	LCC	LPC
SLA	1					
Plant height	-0.386	1				
Seed mass	<i>-0.654</i>	0.522	1			
LNC	0.271	-0.005	-0.402	1		
LCC	-0.101	0.667	0.523	0.097	1	
LPC	-0.078	<i>-0.610</i>	-0.383	0.112	-0.560	1

Table 2. Species-level analysis: ANOVA of responses of plant traits to grazing treatments. Morphological traits analyzed for the 2003-2004 dataset (15 species) were specific leaf area (SLA), plant height, seed mass, leaf dry matter content (LDMC), and leaf area. Chemical and morphological traits analyzed for the 2003 dataset (11 species) were SLA, plant height, seed mass, leaf nitrogen content (LNC), leaf carbon content (LCC), and leaf phosphorus content (LPC).

		2003-2004 dataset													
		Plant height		Seed mass		LDMC		Leaf area							
Source	SLA	F	P	df	F	P	df	F	P						
Treatment	2	18.8	0.003	2	1.53	0.348	2	59.9	<0.0001	2	6.27	0.004	2	2.14	0.135
Plot (within Treatment)	3	0.068	0.977	3	2.26	0.090	3	0.010	0.999	3	0.004	1.00	3	0.010	0.999
Species (within Plot within Treatment)	54	17.7	<0.0001	58	6.20	<0.0001	34	104	<0.0001	54	27.7	<0.0001	54	39.0	<0.0001
Error	226			215			131			226			226		
		2003 dataset													
		Plant height		Seed mass		LNC		LCC		LPC					
Source	SLA	F	P	df	F	P	df	F	P	df	F	P			
Treatment	2	9.22	0.028	2	1.15	0.426	2	55.1	<0.0001	2	8.37	0.055	2	8.00	0.059
Plot (within Treatment)	3	0.051	0.985	3	1.80	0.164	3	0.011	0.998	3	0.151	0.929	3	0.138	0.937
Species (within Plot within Treatment)	46	20.0	<0.0001	38	7.12	<0.0001	30	103	<0.0001	50	11.0	<0.0001	50	2.18	0.005
Error	199			154			117			42			42	5.04	<0.0001

was marginal (Table 1). At the community level, LCC was correlated negatively to SLA and positively to seed mass, while LPC was positively correlated to SLA. LCC was significantly correlated to RDA Axis 1, while there were no significant correlations of leaf chemical functional parameters with Axis 2 (Table 3).

Discussion

LHS strategy scheme

The LHS strategy scheme is based on the assumption that each of the three traits varies widely between species at any given level of the other two, i.e., a degree of independence is assumed to exist between the three traits (Westoby 1998). Correlation between any two of the traits reduces the applicability of the scheme as three-dimensional. Many researchers have found that vegetative traits (e.g., SLA) and regenerative traits (e.g., seed mass) are not usually tightly coupled (e.g., see Weiher et al. 1999). However, two recent tests of the LHS scheme by Lavergne et al. (2003) and Garnier et al. (2004) found a strong positive correlation between plant height and seed mass, thus reducing the number of discriminating traits to two. Our approach to this scheme focused on the community level (Moog et al. 2005), in addition to the species level (Lavergne et al. 2003; Garnier et al. 2004). Therefore, our results also reflect the effect of community structure due to grazing treatments on the average value of each plant trait and on the relationships between traits across communities. At the species level, SLA and seed mass were highly correlated, thus the assumption of Westoby's (1998) theory is not upheld. At the community level, the three LHS parameters (SLA, plant height, and seed mass) combined to form one dimension of variation in the RDA, which basically differentiated between grazed plots and protected plots.

Community functional parameters were weighted according to the species contribution to biomass, so parameter values reflected the trait values of the species making the greatest contribution to total biomass. It can be argued that the response of community functional parameters to protection from grazing may reflect the response of the grasses, which dominate the species list. Indeed, Lavorel et al. (1997) and McIntyre & Lavorel (2001) have shown that different life forms may respond differently to disturbance. However, since we found no evidence of such a "grass effect" in our data

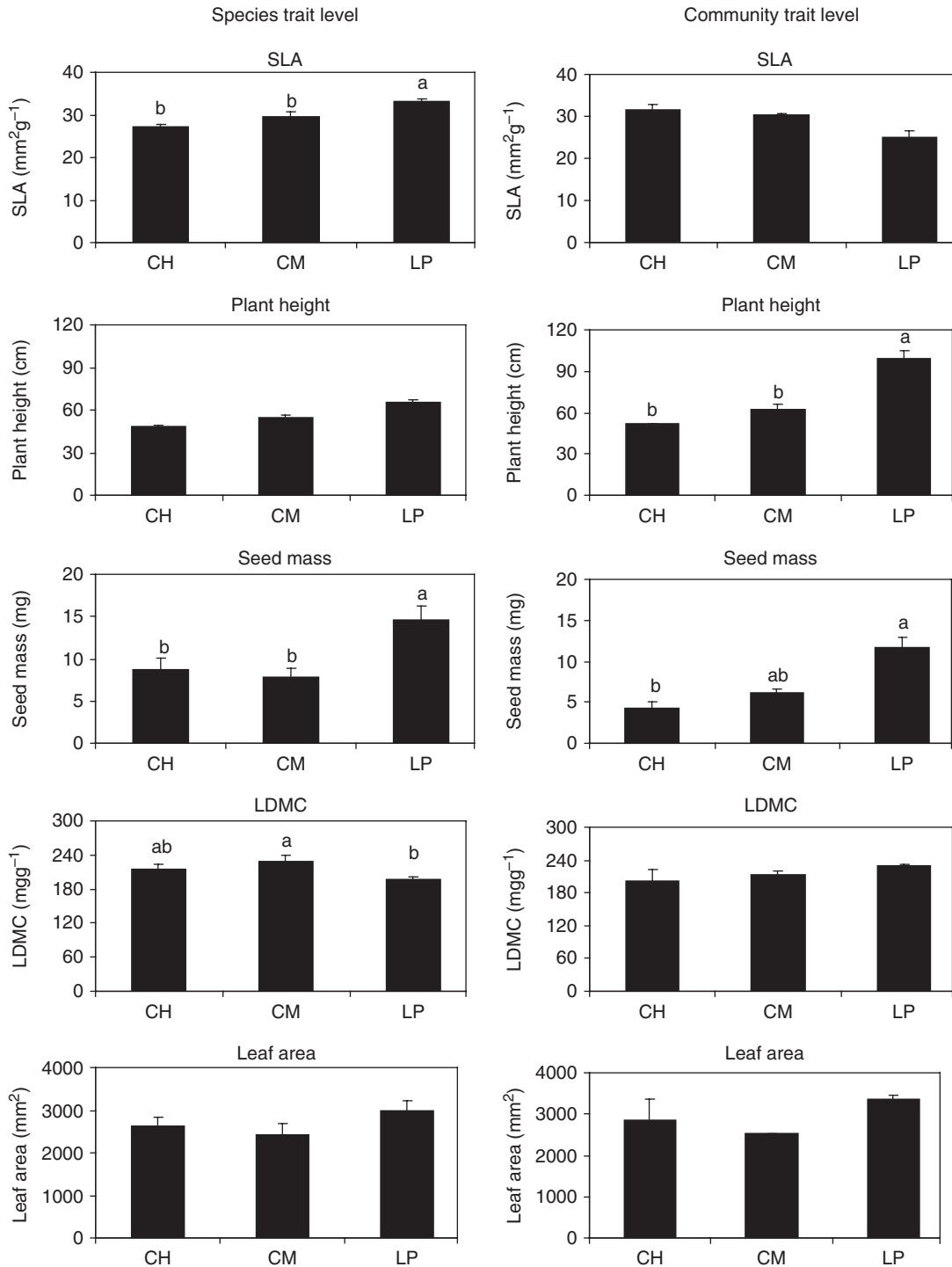


Fig. 1. Response of species traits (left) and community functional parameters (right) to grazing treatments for 15 species fulfilling the 80% biomass threshold. CH – heavy grazing; CM – moderate grazing; LP – long-term protection from grazing. Data pooled for 2003 and 2004. Columns with different lowercase letters are significantly different according to Tukey HSD ($\alpha = 0.05$). Columns are means with standard error.

analysis, we infer that the significant responses of community functional parameters to protection from grazing were generally independent of grass productivity. The only exception was LPC, which,

due to its strongly negative correlation to grass biomass, showed a significant positive response to protection from grazing after we accounted for grass biomass.

Table 3. ANOVA of responses of community functional parameters to grazing treatments. Morphological parameters analyzed for the 2003-2004 dataset (15 species) were specific leaf area (SLA), plant height, seed mass, leaf dry matter content (LDMC), and leaf area. Chemical and morphological parameters analyzed for the 2003 dataset (11 species) were SLA, plant height, seed mass, leaf nitrogen content (LNC), leaf carbon content (LCC), and leaf phosphorus content (LPC).

Source	2003-2004 dataset												
	SLA			Plant height		Seed mass		LDMC		Leaf area			
	<i>df</i>	<i>F</i>	<i>P</i>	<i>F</i>	<i>P</i>	<i>F</i>	<i>P</i>	<i>F</i>	<i>P</i>	<i>F</i>	<i>P</i>		
Treatment	2	5.42	0.101	27.1	0.012	14.2	0.030	0.920	0.488	1.73	0.316		
Error	3												
Source	2003 dataset												
	SLA			Plant height		Seed mass		LNC		LCC		LPC	
	<i>df</i>	<i>F</i>	<i>P</i>	<i>F</i>	<i>P</i>	<i>F</i>	<i>P</i>	<i>F</i>	<i>P</i>	<i>F</i>	<i>P</i>	<i>F</i>	<i>P</i>
Treatment	2	62.3	0.004	168	0.001	28.7	0.011	8.91	0.055	39.9	0.007	0.709	0.560
Error	3												

Table 4. Spearman correlation coefficients of community functional parameters with each other, with RDA axes, and with grazing intensity. Morphological parameters analyzed for the 2003-2004 dataset (15 species) were specific leaf area (SLA), plant height, seed mass, leaf dry matter content (LDMC), and leaf area. Chemical and morphological parameters analyzed for the 2003 dataset (11 species) were SLA, plant height, seed mass, leaf nitrogen content (LNC), leaf carbon content (LCC), and leaf phosphorus content (LPC). Bold values are significant ($\alpha = 0.05$); italicized values are marginally significant ($\alpha = 0.1$). NA – not applicable. ¹SLA, plant height, seed mass; ²SLA, plant height, seed mass, LDMC, leaf area.

	2003-2004 dataset					
	SLA	Plant height	Seed mass	LDMC	Leaf area	
SLA	1					
Plant height	-0.943	1				
Seed mass	-0.943	1.000	1			
LDMC	-0.943	0.829	0.829	1		
Leaf area	-0.257	0.314	0.314	0.143	1	
Grazing intensity	0.837	-0.956	-0.956	-0.717	-0.478	
Axis 1: LHS ¹	0.829	-0.923	-0.911	NA	NA	
Axis 2: LHS	-0.551	0.358	0.412	NA	NA	
Axis 1: 5 traits ²	0.829	-0.923	-0.911	-0.610	-0.444	
Axis 2: 5 traits	0.360	-0.062	-0.199	-0.610	0.732	
	2003 dataset					
	SLA	Plant height	Seed mass	LNC	LCC	LPC
SLA	1					
Plant height	-0.600	1				
Seed mass	-0.714	0.943	1			
LNC	-0.714	0.486	0.429	1		
LCC	-0.943	0.657	0.829	0.600	1	
LPC	0.829	-0.0857	-0.314	-0.429	-0.771	1
Grazing intensity	0.598	-0.956	-0.956	-0.478	-0.717	0.120
Axis 1	<i>0.795</i>	-0.926	-0.930	-0.376	-0.815	0.164
Axis 2	0.603	-0.354	-0.342	-0.705	-0.579	0.764

A high value of the SLA community functional parameter was associated with grazed plots, which are dominated by species with a high SLA, e.g., short annual grasses (see Table S2). With decreased grazing intensity and protection from grazing, SLA decreases, as fast-growing species typical of heavily grazed environments (Westoby 1999) are replaced by slower-growing species (e.g., the perennial

H. bulbosum), which conserve resources in basal storage organs as succession proceeds (Moog et al. 2005). Conversely, plant height and seed mass both increased under protection from grazing. Plant height generally increases with release from uniform high-intensity grazing (Westoby 1999), as height is advantageous in conditions of strong competition for light (Noy-Meir et al. 1989; Moog et al. 2005).

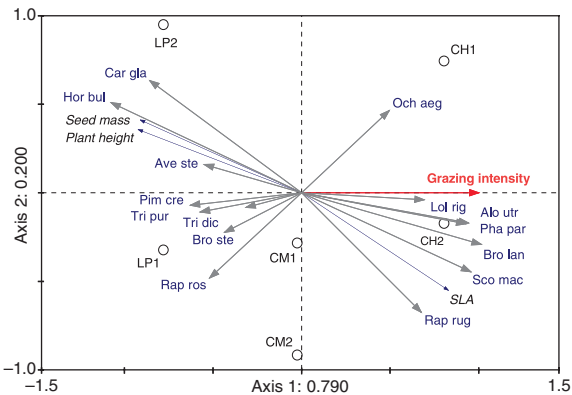


Fig. 2. Community-level analysis: RDA of community functional parameters of LHS traits (specific leaf area (SLA), plant height, seed mass) using pooled data for 2003 and 2004. Eigenvalues are indicated on the axes. CH1, CH2 – high grazing intensity, CM1, CM2 – moderate grazing intensity, LP1, LP2 – long-term protection from grazing. Species were superimposed onto the RDA. Species abbreviations: Alo utr (*Alopecurus utriculatus*), Ave ste (*Avena sterilis*), Bro lan (*Bromus lanceolatus*), Bro ste (*B. sterilis*), Car gla (*Carthamus glaucus*), Hor bul (*Hordeum bulbosum*), Lol rig (*Lolium rigidum*), Och aeg (*Ochthodium aegyptiacum*), Pha par (*Phalaris paradoxa*), Pim cre (*Pimpinella cretica*), Rap ros (*Raphanus rostratus*), Rap rug (*Rapistrum rugosum*), Sco mac (*Scolymus maculatus*), Tri pur (*Trifolium purpureum*), Tri dic (*Triticum dicoccoides*).

Similarly, seed mass usually increases since: (a) taller species have the potential to produce larger seeds (Leishman et al. 1995; Osem et al. 2006; but see Aarssen 2005), and (b) large seeds are advantageous for germination in shade (Westoby et al. 1996) and for penetration of the litter layer (Sternberg et al. 2003; Moog et al. 2005), i.e., the ability to germinate in suboptimal conditions dominates over maximizing opportunities for seedling establishment (Westoby 1999).

Grazing intensity explained less variation in community functional parameters when we added traits to the core set of LHS traits (59% and 53% compared to 79%). This explained variation was not significant, due to the small number of plots used in the analysis; however, we can still draw some insights from the results. The additional community functional parameters (LDMC, leaf area, LNC, LCC, LPC) were not significantly correlated to grazing intensity, and, with the exception of LCC, correlations to the ordination axes and responses to protection from grazing were non-significant. We may thus conclude that the LHS traits are most appropriate for describing variation in this grassland community with a high dominance of annuals, con-

firmed Westoby's (1998) choice of traits and the conclusions of Weiher et al. (1999).

Based on community-level correlations, we can divide the studied traits into two groups that are located at opposite ends of the grazing gradient. The first comprises LDMC, plant height, seed mass, and LCC, associated with protected plots, and the second comprises SLA, associated with grazed plots. SLA was positively correlated to LPC; however, this relationship is probably due to the strong correlation between LPC and grass biomass. The negative correlation of SLA with all other traits reflects the trade-off between growth and reproductive allocation (Suding et al. 2003). In frequently disturbed, grazed plots, species have rapid growth rates, i.e., high SLA, and decreased investment in supportive tissues, resulting in thinner or less dense leaf tissues (Garnier et al. 1997; Wright & Westoby 1999), and lower LDMC (Suding et al. 2003) and LCC. Plants tend to be smaller, and invest less in individual seed reserves, producing many small seeds (Suding et al. 2003; Navas & Moreau-Richard 2005). Conversely, species with a low SLA are larger and taller, with slower growth rates and increased investment in structural components (i.e., higher LDMC and LCC) and seed reserves (Navas & Moreau-Richard 2005). Such species were common in ungrazed plots, where conditions of low light, higher litter accumulation, and the absence of grazing benefited taller, larger-seeded species with slower growth rates (Marañón & Grubb 1993; Moog et al. 2005).

The community-level approach

In this research we used a community-level approach to test the LHS scheme and to investigate trait relationships in a Mediterranean grassland dominated by annuals. Plant trait relationships within this community support those found in a broad range of plant communities (Westoby et al. 1996; Garnier et al. 1997; Wright & Westoby 1999; Suding et al. 2003; Moog et al. 2005; Navas & Moreau-Richard 2005).

We found some significant relationships between community functional parameters, but at the species level trait relationships were largely non-significant. The lack of significant correlations at the species level arises from the large overlap in species composition between treatments, with most common species being present in all treatments. Differences between treatments reflected changes in dominance, rather than species identity. Significant correlations observed at the community level reflected the trait correlations characteristic of the

dominant species. Thus, community functional parameters proved to be the preferred measure for understanding the dynamics of trait relationships and responses to protection from grazing. Community-level dynamics are important for understanding ecosystem-level responses (e.g., productivity, nutrient cycling) to protection from grazing. We found that community functional parameters of the three key plant traits in the LHS model captured most of the variation in species composition between the experimental treatments.

Despite the relative success of the LHS model at the community level, and the significant correlations between grazing intensity and LHS parameters, the model could not discriminate between the applied levels of grazing intensity. This was due to the similarity in species composition between the two grazing treatments (data not shown), despite the large difference in consumption of plant material (65% for CH compared to 31% for CM). This high similarity precludes screening of additional plant functional traits to improve the model's success, since differences in trait relationships always depend on variations in species composition between treatments. In contrast, we can conclude that the model is successful at discriminating between grazed and ungrazed treatments due to the lower similarity between communities in grazed versus ungrazed plots. Nevertheless, this study shows that high species similarity may be problematic when using aggregate community analysis if it is not associated with large differences in species frequencies between experimental treatments.

Acknowledgements. This research was carried out as part of the EU-funded project VISTA (Vulnerability of Ecosystem Services to Land-Use Change in Traditional Agricultural Landscapes), EVK2-2002-00356. The study was part of the PhD thesis of C.G. We thank Imanuel Noy-Meir, Sandra Lavorel, Phil Lambdon, and an anonymous referee for constructive comments and advice on earlier versions of the manuscript. Thanks also to Hillary Voet for providing statistical advice. Particular acknowledgement is made to Ingolf Kühn for his dedication and commitment to the success of this paper. We also thank the many students who assisted in data collection and processing.

References

Aarssen, L.W. 2005. Why don't bigger plants have proportionately bigger seeds? *Oikos* 111: 199–207.
Cornelissen, J.H.C., Lavorel, S., Garnier, E., Diaz, S., Buchmann, N., Gurvich, D.E., Reich, P.B., ter Steege,

H., Morgan, H.D., van der Heijden, M.G.A., Pausas, J.G. & Poorter, H. 2003. A handbook of protocols for standardized and easy measurement of plant functional traits worldwide. *Australian Journal of Botany* 51: 335–380.

Diaz, S., Lavorel, S., McIntyre, S., Falczuk, V., Casanoves, F., Milchunas, D.G., Skarpe, C., Rusch, G., Sternberg, M., Noy-Meir, I., Landsberg, J., Zhang, W., Clark, H. & Campbell, B.D. 2006. Plant trait responses to grazing – a global synthesis. *Global Change Biology* 12: 1–29.

Feinbrun-Dothan, N. & Danin, A. 1991. *Analytical flora of Eretz-Israel*. Cana Publishers, Jerusalem.

Garnier, E., Cordonnier, P., Guillermin, J.-L. & Sonié, L. 1997. Specific leaf area and leaf nitrogen concentration in annual and perennial grass species growing in Mediterranean old-fields. *Oecologia* 111: 490–498.

Garnier, E., Bellman, A., Navas, M.-L., Roumet, C. & Laurent, G. 2004. The leaf–height–seed plant ecology strategy scheme as applied to species from a Mediterranean old-field succession. In: Arianoutsou & Papanastasis (eds.) *Proceedings 10th MEDECOS Conference, Rhodes, Greece*. Millpress, Rotterdam, NL.

Garnier, E., Lavorel, S., Ansquer, P., Castro, H., Cruz, P., Dolezal, J., Eriksson, O., Fortunel, C., Freitas, H., Golodets, C., Grigulis, K., Jouany, C., Kazakou, E., Kigel, J., Kleyer, M., Lehsten, V., Lepš, J., Meier, T., Pakeman, R., Papadimitriou, M., Papanastasis, V., Quested, H., Quétier, F., Robson, M., Roumet, C., Rusch, G., Skarpe, C., Sternberg, M., Theau, J.-P., Thebault, A., Vile, D. & Zarovali, M. 2007. Assessing the effects of land-use change on plant traits, communities and ecosystem functioning in grasslands: a standardized methodology and lessons from an application to 11 European sites. *Annals of Botany* 99: 967–985.

Grime, J.P. 1974. Vegetation classification by reference to strategies. *Nature* 250: 26–31.

Grime, J.P. 1998. Benefits of plant diversity to ecosystems: immediate, filter and founder effects. *Journal of Ecology* 86: 902–910.

Grime, J.P. 2001. *Plant strategies, vegetation processes and ecosystem properties*. 2nd ed. John Wiley & Sons, Chichester.

Lavergne, S., Garnier, E. & Debussche, M. 2003. Do rock endemic and widespread plant species differ under the Leaf–Height–Seed plant ecology strategy scheme? *Ecology Letters* 6: 398–404.

Lavorel, S., McIntyre, S., Landsberg, J. & Forbes, T.D.A. 1997. Plant functional classifications: from general groups to specific groups based on response to disturbance. *TREE* 12: 474–478.

Leishman, M.R., Westoby, M. & Jurado, E. 1995. Correlates of seed size variation: a comparison among five temperate floras. *Journal of Ecology* 83: 517–529.

Lepš, J. & Šmilauer, P. 2003. *Multivariate analysis of ecological data using CANOCO*. Cambridge University Press, Cambridge, UK.

- Marañón, T. & Grubb, P.J. 1993. Physiological basis and ecological significance of the seed size and relative growth rate relationship in Mediterranean annuals. *Functional Ecology* 7: 591–599.
- McIntyre, S. & Lavorel, S. 2001. Livestock grazing in subtropical pastures: steps in the analysis of attribute response and plant functional types. *Journal of Ecology* 89: 209–226.
- Moog, D., Kahmen, S. & Poschlod, P. 2005. Application of CSR and LHS strategies for the distinction of differently managed grasslands. *Basics of Applied Ecology* 6: 133–143.
- Navas, M.-L. & Moreau-Richard, J. 2005. Can traits predict the competitive response of herbaceous Mediterranean species? *Acta Oecologica* 27: 107–114.
- Noy-Meir, I., Gutman, M. & Kaplan, Y. 1989. Responses of Mediterranean grassland plants to grazing and protection. *Journal of Ecology* 77: 290–310.
- Osem, Y., Perevolotsky, A. & Kigel, J. 2006. Size traits and site conditions determine changes in seed bank structure caused by grazing exclusion in semiarid annual plant communities. *Ecography* 29: 11–20.
- Pakeman, R.J. & Queded, H.M. 2007. Sampling plant functional traits: what proportion of the species need to be measured? *Applied Vegetation Science* 10: 93–98.
- Raunkiaer, C. 1937. *Plant life forms*. Clarendon Press, Oxford, UK.
- Sternberg, M., Gutman, M., Perevolotsky, A., Ungar, E.D. & Kigel, J. 2000. Vegetation response to grazing management in a Mediterranean herbaceous community: a functional group approach. *Journal of Applied Ecology* 37: 224–237.
- Sternberg, M., Gutman, M., Perevolotsky, A. & Kigel, J. 2003. Effects of grazing on soil seed bank dynamics: an approach with functional groups. *Journal of Vegetation Science* 14: 375–386.
- Suding, K., Nash, Goldberg, D.E. & Hartman, K.M. 2003. Relationships among species traits: separating levels of response and identifying linkages to abundance. *Ecology* 84: 1–16.
- Ter Braak, C.J.F. & Šmilauer, P. 2002a. *CANOCO for windows 4.5*. Microcomputer Power, Ithaca, NY, USA.
- Ter Braak, C.J.F. & Šmilauer, P. 2002b. *CanoDraw for windows 4.0*. Microcomputer Power, Ithaca, NY, USA.
- Violle, C., Navas, M.-L., Vile, D., Kazakou, E., Fortunel, C., Hummel, I. & Garnier, E. 2007. Let the concept of trait be functional!. *Oikos* 116: 882–892.
- Weiher, E., van der Werf, A., Thompson, K., Roderick, M., Garnier, E. & Eriksson, O. 1999. Challenging theophrastus: a common core list of plant traits for functional ecology. *Journal of Vegetation Science* 10: 609–620.
- Westoby, M. 1998. A leaf–height–seed (LHS) plant ecology strategy scheme. *Plant Soil* 199: 213–227.
- Westoby, M. 1999. The LHS strategy scheme in relation to grazing and fire. In: Eldridge, D. & Freudenberger, D. (eds.) *Proceedings of the VI International Rangelands Congress, Australia*. Vol. 2, pp. 893–896. International Rangeland Congress, St Ives, AU.
- Westoby, M., Leishmann, M. & Lord, J. 1996. Comparative ecology of seed size and dispersal. *Philosophical Transactions of the Royal Society of London B Biological Sciences* 351: 1309–1318.
- Westoby, M., Falster, D.S., Moles, A.T., Vesk, P.A. & Wright, I.J. 2002. Plant ecological strategies: some leading dimensions of variation between species. *Annual Review of Ecology and Systematics* 33: 125–159.
- Wright, I.J. & Westoby, M. 1999. Differences in seedling growth behaviour among species: trait correlations across species, and trait shifts along nutrient compared to rainfall gradients. *Journal of Ecology* 87: 85–97.
- Zohary, M. 1973. *Geobotanical foundations of the Middle East*. Vols. 1 and 2, Gustav Fischer Verlag, Stuttgart - Swets and Zeitlinger, Amsterdam, NL.

Supporting Information

Additional Supporting Information may be found in the online version of this article:

Table S1. Species fulfilling the 80% biomass requirement for 2003 and 2004.

Table S2. Percentage biomass values for species examined in each of the experimental plots for 2003 and 2004.

Please note: Blackwell Publishing are not responsible for the content or functionality of any supporting materials supplied by the authors. Any queries (other than missing material) should be directed to the corresponding author for the article.

Received 6 February 2007;

Accepted 3 March 2008.

Co-ordinating Editor: I. Kühn.