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Assessment of grazing effects on mediterranean shrubs *Phillyrea latifolia* L. and *Cistus monspeliensis* L. with developmental instability and fractal dimension

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Abstract

In this study we used developmental instability expressed as translational and fluctuating asymmetries as well as fractal dimension in order to assess the effects of different levels of grazing pressure by goats on two dominant species of Mediterranean shrublands, i.e. *Phillyrea latifolia* L., a palatable, and *Cistus monspeliensis* L., an unpalatable shrub. Treatments included light, moderate and heavy grazing pressure. For the former species, an additional ungrazed treatment was used as a control. Measurements were carried out in thirty shrubs of each species in each treatment. For *Phillyrea*, the fluctuating asymmetry analysis revealed higher asymmetry of leaves in the grazed treatments than in the control. The lower parts (below 1.5m height) of *Phillyrea* shrubs, reached by goats, also attained larger values of asymmetry in comparison to the upper parts (above 1.5m height) of the shrubs, not reached by goats. Moreover, fractal dimension of branches was lower in the grazed treatments in comparison to the ungrazed ones, as well as in the lower parts compared to the upper parts of the shrubs. However, the results of translational asymmetry analysis showed that grazed shrubs were developmentally more stable than the ungrazed ones. These results suggest that *Phillyrea* is very tolerant to grazing; its upper part can be maintained in a non - stressful situation even when its lower part is overgrazed. For *Cistus*, the results of translational asymmetry revealed that grazing did not affect its developmental stability. The effects of grazing on fractal dimension declined under moderate grazing situations. This indicates that *Cistus* is developmentally stable both in well preserved and in heavily disturbed sites. It is concluded that developmental instability and fractal dimension are useful tools in assessing the differential response of Mediterranean key species to grazing impact.

Introduction

Mediterranean shrublands are ecosystems with a long history of grazing by domestic animals. Light grazing has no profound affects on vegetation and soils, while moderate grazing is considered to be essential for maintaining species diversity and ecosystem stability (McNaughton, 1983; Noy-Meir, 1998). Overgrazing, on the contrary, is the major cause of desertification, especially where poor and erodible soils are found (Arianoutsou - Faraggitaki, 1985; Papanastasis et al., 1990; Margaris and Koutsidou, 1998). In such shrublands, dominant shrub species may be used as key-indicators of the grazing impact on vegetation.

Developmental instability and fractal dimension as measures of stress can be used to resolve the controversy over whether light to moderate grazing enhances the fitness of the grazed plants (McNaughton, 1977; Collins, 1987; Escós et al. 1997).

Developmental stability is the ability of an individual to produce an invariant phenotype under particular environmental conditions (Møller and Swaddle, 1997). It also represents its ability to buffer random environmental perturbations, such as negative feedback mechanisms, hormonal regulation and central neural regulation during development (Mather, 1953; Palmer, 1994). These processes may have a genetic basis, meaning that some genotypes have a superior developmental precision under certain conditions (Mather, 1953; Perfectti and Camacho, 1999), but this is still being discussed (Clarke, 1993; Markow and Clarke, 1997). Developmental instability, on the other hand, does not have a heritable basis and represents the effects of random environmental disturbances i.e. herbivory, competition, pollution or even high altitude (Waddington, 1957; Martel et al., 1999). It is reflected by exaggerated intra-individual variation in repeated traits and patterns.

Several methods have been used in order to estimate developmental instability by measuring the within-plant variation such as: frequency of phenodeviants; deviations from radial, spiral, translational and bilateral symmetries (i.e. fluctuating asymmetry); and fractal dimensions of plant structures (Mather, 1953; Palmer and Strobeck, 1986; Freeman et al., 1993; Graham et al., 1993; Møller and Swaddle, 1997; Alados et al., 1998). According to Alados et al. (1999), any phenotypic trait can be used in order to measure developmental instability, provided that the phenotype produced in the absence of stress is known. Translational asymmetry, represented by the error in the regular and consistent variation of internode space with order of nodes (Freeman et al., 1993, Alados et al., 1999) has been successfully used in several studies for the estimation of developmental instability in plants (Graham et al., 1993; Freeman et al., 1993; Escós et al., 1997). Fluctuating asymmetry is also widely used in measuring developmental instability for bilaterally and radially symmetrical traits (Palmer, 1994; Møller and Shykoff, 1999). It is defined as the pattern of bilateral variation in a sample of individuals where the mean of right minus left (R-L) side difference is zero and variation is normally distributed about that mean (Palmer, 1994).

On the other hand, due to the modular nature of plants allometric relations follow power laws, i.e. the same pattern is repeated over many scales. Power law scaling results from self-similarity (Alados et al., 1998), that is typical of fractal structures and common in plants (Lindenmayer, 1968; Freeman et al., 1993). According to Freeman et al. (1993), a modular unit produced this year in woody plants is similar in morphology to the modular unit produced last year.

In order to analyze these self-similar structures, fractal analysis and more specifically fractal dimension can be used. Fractal dimension is a measure of the degree to which space is filled and thus how efficiently plants interact with their environment. Complex plant structures have higher fractal dimension, which means that they exchange more substances with their environment than less complex ones (Alados et al., 1999).

In this paper, we studied the effect of grazing on developmental instability and fractal dimension of two dominant shrub species in a Mediterranean shrubland, one palatable and another unpalatable, so that grazing impact caused by pastoral management is assessed. Moreover, we tried to prove by measuring the fluctuating asymmetry and the fractal dimension of the upper, not reached and the lower, reached by grazing animals, parts of shrubs, that developmental instability is the result of external environmental disturbances without genetic influence, since both parts of plants have the same genotype.

Materials and methods

The study area is located in the village community of Sykia at the south part of the Sithonia peninsula in Chalkidiki, northern Greece (longitude: 23°54’, latitude: 40°00’). The climate is semi-arid to sub-humid Mediterranean with an annual average rainfall of 590mm. The bedrock consists of mainly metamorphic rocks of the Mesozoic era while there are also sites covered by sedimentary rocks. The natural vegetation are maquis-type shrublands dominated by the shrub species *Cistus monspeliensis* L., *Pistacia lentiscus* L., *Phillyrea latifolia* L., *Olea europaea* L., and *Quercus coccifera* L.. In addition, several herbaceous species are present including grasses and broadleaved species. The area is mainly grazed by goats. Goat flocks are corralled at night in permanent sheds distributed throughout the village community territory and graze freely around them the whole year round to a distance of 1-2 km. Areas closer to sheds are more frequently visited and therefore heavier grazed than the ones away from them.

In early spring of 2000, a representative shed was selected where 300 goats were housed. After locating the grazing territory of the flock and studying its grazing behavior, three homocentric zones were established around the shed. The inner zone was grazed with a stocking rate of 8.23 goats/ha/yr and characterized as heavily grazed area, the middle zone with 2.63 goats/ha/yr and characterized as moderately grazed and the outer zone with 0.28 goats/ha/yr and characterized as lightly grazed area. In addition, an ungrazed area located far from the shed, but within the village territory, was selected as a control.

Among the dominant species, *Phillyrea* was the most palatable shrub, readily consumed by goats, while *Cistus* was unpalatable. The majority of *Phillyrea* shrubs exceeded 1.5m height while of *Cistus* were less than this height.

In May 2000, 30 adult plants of *Phillyrea* and 30 ones of *Cistus* were randomly selected in each of the three treatments for measurements and in the control. For the latter species, no such shrubs were taken in the control treatment because it was thought that they were not necessary since the species is not preferred by goats.

Translational asymmetry

For translational asymmetry, two well-developed and undamaged shoots from each shrub were randomly collected, pressed and dried. In *Phillyrea*, these shoots were taken from the upper part (above 1.5m height) of shrubs that was not reached, and, therefore, not grazed by goats while in *Cistus*, from the middle strata of the plants.

In each shoot, the internode length was measured from the bottom to the top using an electronic calliper. The relationship between internode length and node order was estimated by the equation (Alados et al., 1994; 1998; Escos et al., 1997):

$$L = KN^{a}e^{-bN}$$

where L is the internode length, N is the node order from the base of the shoot to its top, e is the natural base and k, a, b, fitted constants (Figure 1). For evaluating the accuracy of curve fitting we calculated the coefficient of determination (R^2), the standard error of regression (S_{yx}), and the standard errors of the parameters a (S_a), and b (S_b). Because the number of internodes varied among shoots, we used R^2 adjusted to the degrees of freedom. Finally, we calculated $S_{yx}/\bar{y}$ (Zar, 1984) because S_{yx} is related to the average internode length.

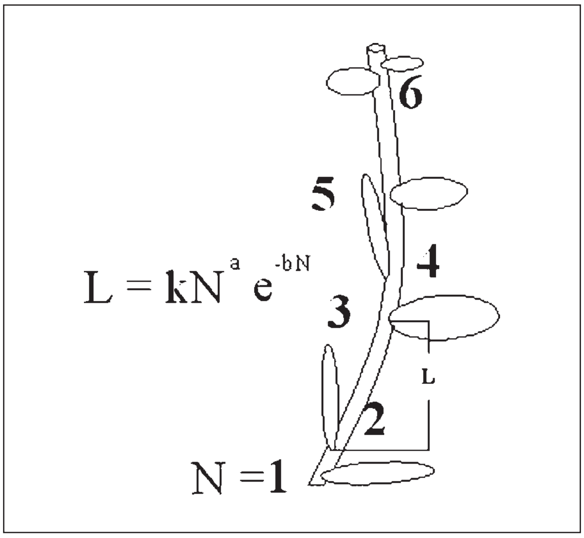

Figure 1. A diagrammatic representation of a shoot with its internode length (L) and node order (N).

In order to estimate the effect of different levels of grazing pressure on the accuracy of curve fitting, a mixed model analysis of covariance was used (model III GLM, SAS) with grazing as a fixed effect factor, the shoot nested into individuals, and the neperian logarithm of shoot diameter as a covariate. Comparisons of means between treatments were made with Tukey's studentized range test.

Fluctuating asymmetry

Fluctuating asymmetry was measured only in *Phillyrea*. More specifically, five leaves were collected both from the upper part (above 1.5m height) not reached by goats, and from the lower part (below 1.5m height) reached by the animals, of each *Phillyrea* shrub. Leaves were selected from intermediate shoot positions to reduce variability.

Leaf measurements were performed by using the OPTIMAS 6.2 image processing. Images were taken with a SONY CV-M90 color camera (512-512 pixels) with a macro objective and a tripod from a fixed focal length. More specifically, we measured the width of the right (R) and left (L) halves of the blade of each leaf at its middle, between petiole insertion and the tip.

In order to use fluctuating asymmetry in developmental instability studies we must ensure the absence of antisymmetry (departures from normality of the frequency distribution of L-R difference) and directional asymmetry (one side is significantly larger than the other on the average) (Palmer and Strobeck, 1992). Both these asymmetries indicate that some portion of the between sides variation of a particular trait may have a genetic basis and not be a pure product of developmental instability (Palmer, 1994). For antisymmetry, we performed normality tests of the distribution of the signed difference (L-R) that included a t-test and the use of the Wilk-Shapiro index (Shapiro et al., 1968) as well as skewness and kurtosis tests (Sokal and Rohlf, 1995). For directional asymmetry, we used a mixed model of two-way analysis of variance. In this analysis, side was the fixed effect factor, representing directional asymmetry; individual leaves were the random factor, assessing leaf size and shape variation among them; the side-by-individual interaction term represented fluctuating asymmetry; and finally the error term referred to measurement variation and represented the measurement error.

Subsequently, a mixed model of two-way analysis of variance was used to examine the effects of different levels of grazing pressure on mean leaf width. The need to correct the size dependence of fluctuating asymmetry was also checked according to Palmer (1994).

This was done with a linear regression of |L-R| to (L+R)/2. In case that this regression revealed that we had a size scaling of fluctuating asymmetry, we removed it by using a Box-Cox transformation of the log (L/R) (CTFA) (Swaddle et al. 1994), and the relative fluctuating asymmetry index (RFA) as the absolute value of left minus right divided by the average ((L+R)/2) (Palmer and Strobeck, 1986). In addition, the FA10 index of Palmer (1994) was also used. It is the only index that provides an estimate of the between sides variance after removing the effects of measurement error. After that, a Bartlett's test was performed to check the statistical significance of the differences. Grazing effects on fluctuating asymmetry was estimated by a mixed model analysis of variance (model III GLM, SAS) as well as with a nested analysis of variance where grazing was the fixed effect factor and location (above and below grazing level) was nested within treatment to detect the effects of different leaf location.

Fractal dimension

For fractal analysis, 5 year-old branches were selected from each shrub and photographed, at a fixed distance, focusing at the center of the plant. For *Phillyrea* in particular, branches were obtained from two locations on the shrubs, namely from the upper part (above 1.5 m height) and the lower parts (0-1.5 m) (Figure 2), while for *Cistus* only one branch was selected from the middle strata of the canopy.

Images taken by the digitized camera were captured with Adobe Photoshop 5.5 with resolution of 3072x2048 pixels. Subsequently, they were processed in a window size of 1024x1024 pixels, filtered, converted to grayscale and adjusted to the same intensity. By using a special computer program developed by J. Escos (personal communication) we calculated the fractal dimension (D_I) of the branching system of *Phillyrea* and *Cistus*. This parameter is a generalization of the capacity dimension (D_C) but it also takes into account the relative probability of the cubes used to cover the set (Farmer et al., 1983). It is given by:

$$D_I = \lim_{\varepsilon \to 0} [I_{(\varepsilon)} / \ln(1/\varepsilon)]$$

where $$I = \sum_{i=1}^{N(\varepsilon)} p_i \ln p_i \text{ and } p_i = x_i / \sum x_i$$

and xi is the number of occupied pixels for each box of size i (Alados et al., 1999).

In addition, we estimated the evenness index that measures the non-uniformity of a structure and serves to discriminate objects that have the same fractal dimension (Smith et al., 1996). It is given by:

$$\overline{J_{(\varepsilon)}} = \sum_{i=1}^{N(\varepsilon)} p_i \ln p_i / \ln N(\varepsilon)$$

In order to estimate the effect of different levels of grazing pressure on the information indices D_I and $\overline{J_{(\varepsilon)}}$, one-way analysis of variance was applied for *Cistus* and a two-way one for *Phillyrea* data.

Results

Phillyrea latifolia

Translational asymmetry

Shoot length varied significantly among treatments (Table 1). All grazed treatments had significantly longer shoots than the control. This difference was highest in the heavily grazed treatment and lowest in the moderately grazed one.

The arrangement of leaves on the shoot followed the equation $L=12.64\, N^{1.97}e^{-0.8N}$, averaged for 238 samples. The intercept of the equation (lnk) was significantly increased by grazing treatments. Grazed plants presented larger intercept, i.e. larger internode length at the base of the shoot than the control. Scaling parameters a and b were not statistically significant (Table 1). These results suggest that the plants followed the same growth rate in all treatments. Also the standard error of the parameters a and b (Sa and Sb) did not produce any significant differences.

The $S_{yx}/\overline{y}$ of the regression was significantly higher in the control than in the grazed treatments (Table 1), indicating that grazed plants, no matter at what stocking rate, were developmentally stable compared to the ungrazed ones. The R^2_{adj} did not differ significantly between the treatments.

Figure 2. Two five-year old branches of *Phillyrea latifolia* collected from the same shrub, one from the lower part of the plant (left), which was reached by the goats, and the other from the upper part (right), that was not reached by the animals.

Table 1. Means ± se and F values of the mixed model analysis of covariance for *Phillyrea latifolia*. Dependent variables are the scale asymmetry parameters of the relation between internode length and node order (Sa, Sb, R^2adj and Syx/$\overline{y}$), and equation parameters a, b and ln*k* were derived from the equation $L = kN^a e^{-bN.}$

Variables	**Ungrazed**	**Grazed treatments**			**$F_{3,174}$**	**P**
		Light	**Moderate**	**Heavy**		
Length ±shoot)	40.058 ± 16.98b[1]	56.633 ± 24.61a	50.833 ± 23.76a	58.655 ± 23.23a	8.45	0.0001***
a	2.096 ± 1.277	2.056 ± 0.833	1.870 ± 0.839	1.836 ± 0.824	0.70	0.555
b	-0.829 ± 0.514	-0.860 ± 0.347	-0.754 ± 0.521	-0.754 ± 0.287	0.37	0.772
Sa	1.181 ± 0.448	1.079 ± 0.504	1.076 ± 0.454	0.993 ± 0.431	1.22	0.303
Sb	0.475 ± 0.217	0.425 ± 0.222	0.425 ± 0.229	0.374 ± 0.185	1.55	0.202
R^2_{adj}	0.309 ± 0.404	0.409 ± 0.423	0.386 ± 0.385	0.404 ± 0.376	0.94	0.423
Syx/$\overline{y}$	0.206 ± 0.082a	0.167 ± 0.079b	0.193 ± 0.078b	0.169 ± 0.075b	3.17	0.026*
ln*k*	2.261 ± 0.629b	2.699 ± 0.538a	2.556 ± 0.507a	2.633 ± 0.448a	8.38	0.0001***

[1] Means with the same letter within the same variable are not significantly different at 0.05 (*) and 0.001 (***) levels of significance

Fluctuating asymmetry

Normality tests showed that our data did not present antisymmetry. More specifically, the t-test revealed that the signed difference between left and right width of *Phillyrea* leaves, averaged for 1189 of them, did not differ significantly from zero (-0.80, P=0.420). The distribution of L-R difference did not depart from normality since the Wilk-Shapiro statistical index (Shapiro et al., 1968) was 0.99 with P=0.6. Finally, the frequency distribution of L-R difference did not skew ($g_1 = -0.02$, $sg_1 = 0.07$, $tg_1 = 0.29 < t_{0.05} = 1.92$ NS) and was leptokurtic ($g_2 = 0.66$, $sg_2 = 0.14$, $tg_2 = 4.67 > t_{0.05} = 1.92$ S), meaning that antisymmetry was not present (Palmer, 1994; Sokal and Rohlf, 1995).

As far as the directional asymmetry is concerned, the mixed model of two-way analysis of variance showed that this was not significant (Table 2). The side by individual interaction thogh was significant and the measurement error was low, thus resulting in significant fluctuating asymmetry. The ratio of the trait size variation due to measurement error was 0.29%. The proportion of fluctuating asymmetry variation due to measurement error was 8.6%.

Table 2. Variance components in a mixed model of two-way ANOVA (side x individual leaves) for *Phillyrea latifolia*.

Source of variation	**df**	**Mean squares**	**F**	**P**
Sides	1	0.220	2.26	0.1332
Individual leaves	1280	2.868	29.28	0.0001***
Side x individual	1279	0.096	11.56	0.0001***
Error	2195	0.008		

***P<0.001.

Mean leaf side width ((L+R)/2) differed significantly among treatments (Table 3). Also, the slope of the regression between absolute fluctuating asymmetry (|L-R|) and average leaf side width ((L+R)/2) indicated significant size scaling for the lightly and heavily grazed areas as well as for the control. The fluctuating asymmetry indices and the results of their analyses are also shown on Table 3. More specifically, RFA, CTFA and FA10 produced significant differences between treatments. The control had significantly lower values of these indices in comparison with the grazed treatments.

The effects of the different levels of grazing pressure and leaf location on the variation of fluctuating asymmetry are shown in Figure 3. The comparison between the upper part of the plants, which is not reached by goats, and the lower part, which can be reached by the animals, revealed that the lower part had smaller leaf width ($F_{4,1181}$=12.47 P<0.001) but larger fluctuating asymmetry ($F_{4,1181}$ values were 4.74, P<0.001 for RFA, and 4.57, P<0.001 for CTFA). In the control treatment, no statistically significant differences were found between the upper and the lower part of the shrubs as it was expected.

Fractal dimension

The fractal dimension (D_1) of the branch structu-

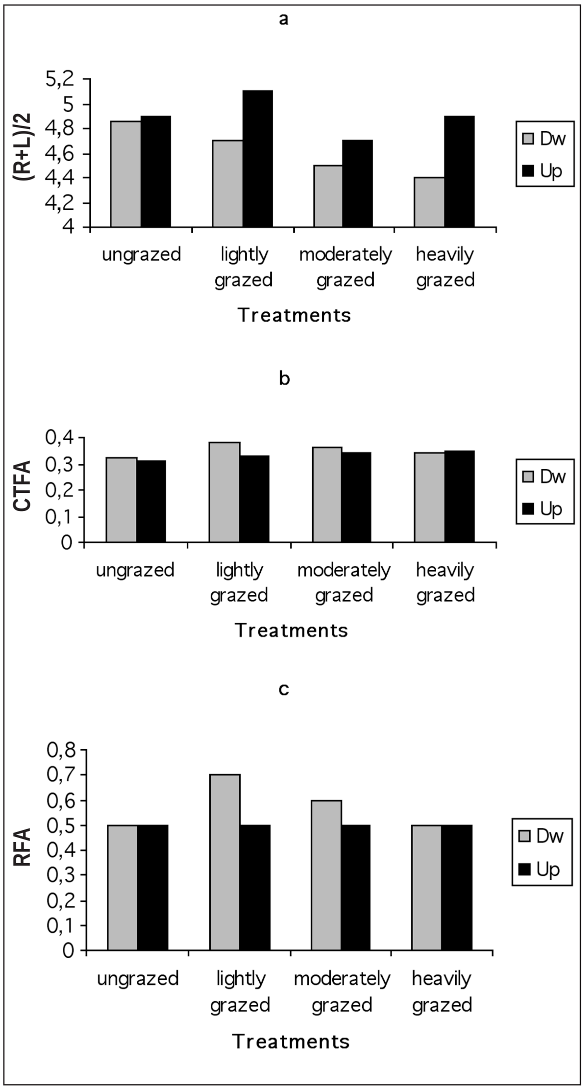

Figure 3. Fluctuating asymmetry indices of the lower (Dw) and upper (Up) parts of *Phillyrea latifolia* shrubs within each grazing treatment **a.** Mean values of the average trait size (L+R)/2 **b.** Box-Cox transformation of log |(L/R)| (CTFA) and **c.** Relative fluctuating asymmetry (RFA) of |L-R|/(L-R)/2.

re of the *Phillyrea* shrubs under different levels of grazing pressure, irrespectively of the location of the branches on the shrub, revealed that the ungrazed plants had significantly higher fractal dimension than the grazed ones ($F_{3,177}$=18.64, P<0.0001) (Table 4). Similar results were also produced for $\overline{J_{(\varepsilon)}}$ which attained its highest value in the control area ($F_{3,177}$=16.63, P<0.0001), where the branches were apparently more uniformly distributed than in the grazed treatments.

On the other hand, significant differences were found between the upper (ungrazed) and the lower (grazed) part of *Phillyrea* shrubs in the information indices D_I, and $\overline{J_{(\varepsilon)}}$ (Table 5).The upper branches showed greater space occupancy ($F_{1,116}$= 1119.77, P<0.001) and a more uniform distribution ($F_{1,116}$=1568.18, P<0.001) of their shoots. Moreover, the interaction between grazing and location of the branches on the shrubs was also statistically significant ($F_{3,232}$ values were 67.52, P<0.0001 for D_I and 111.81, P<0.001 $\overline{J_{(\varepsilon)}}$ (Table 6). In the ungrazed treatment, Duncan's test revealed that the D_I values did not differ between the upper and lower parts except for $\overline{J_{(\varepsilon)}}$ that differed significantly between the two locations.

Cistus monspeliensis

Translational asymmetry

For *Cistus*, shoot length did not vary significantly between the different levels of grazing pressure, as expected (Table 7).

The leaf arrangement on the shoots fitted the equation $L=8.39\ N^{0.97}e^{-0.24N}$, averaged for 179 samples. The intercept of the equation (lnk) was significantly affected by the different grazing treatments. Lightly and moderately grazed plants presented larger intercept, i.e. larger internode length at the base of the shoot than the heavily grazed ones. Scaling parameter a was significantly different between the grazing treatments and attained its highest value in the heavily grazed area meaning that the plants started growing earlier in the season than in the other treatments. On the contrary, no statistically significant differences were found for the scaling parameter b suggesting that the plants in all grazing treatments presented the same inhibitory growth ratio.

The R^2_{adj} and the standard error of the regression after the normalization by the average internode size ($S_{yx}/\overline{y}$) were not significantly affected by grazing pressure.

Fractal dimension

D_I, and $\overline{J_{(\varepsilon)}}$ in *Cistus* were significantly different among the grazing treatments ($F_{2,87}$ values were 8.65 P<0.001 for D_I, 5.36 P<0.01 for $\overline{J_{(\varepsilon)}}$) (Table 8). The plants of the lightly and heavily grazed areas presented greater branch occupancy than shrubs growing in the moderately grazed area. Also branches were more heterogeneously distributed under the moderate grazing conditions as the evenness index attained its lowest value in this treatment.

Table 3. Mean values of the fluctuating asymmetry indices for *Phillyrea latifolia* leaves corresponding to different grazing pressure levels. Means ± se (n) of average leaf size (L+R)/2, slope ± se of the regression between absolute asymmetry vs. average leaf side length. Relative fluctuating asymmetry (RFA) is |L-R|/(L-R)/2 and Box-Cox transformation of log (L/R) is the CTFA. FA10 values from Palmer (1994) (MSm is the measurement error, ${}_{i}^{2}$ indicates the fluctuating asymmetry and df are the degrees of freedom after partitioning out the measurement error).

Treatments	**n**	**(L+R)/2**		**RFA**	**CTFA**	**FA10**		
		x ± se	**slope ±se**	**x ± se**	**x ± se**	**MSm**	${}_{i}^{2}$	**df**
Control	299	4.877 ± 0.05	0.025 ± 0.01^{*1}	0.047 ± 0.04	0.321 ± 0.11	0.00541	0.0276	246
Lightly grazed	300	4.898 ± 0.05	0.054 ± 0.02***	0.056 ± 0.04	0.357 ± 0.12	0.0115	0.0541	243
Moderately grazed	300	4.584 ± 0.05	0.026 ± 0.02	0.056 ± 0.05	0.350 ± 0.12	0.00674	0.0508	262
Heavily grazed	290	4.661 ± 0.06	0.059 ± 0.01***	0.054 ± 0.04	0.349 ± 0.11	0.00876	0.0493	242
$F_{3,1181}$		10.03***		3.11*	5.72***	11.56***		

[1] Means with the same letter within the same variable are not significantly different at 0.05 (*) and 0.001 (***) levels of significance

Table 4. Means of fractal dimension (D_I), and average evenness index $\overline{J(\varepsilon)}$ of *Phillyrea latifolia* in the four grazing treatments.

Variable	**Ungrazed**	**Grazed treatments**		
		Light	**Moderate**	**Heavy**
D_I	1.61a[1]	1.46b	1.48b	1.46b
$\overline{J(\varepsilon)}$	0.61a	0.50b	0.51b	0.49b

[1] Means with the same letter within the same variable are not significantly different at the 0.001 level of significance.

Table 5. Means of information dimension (D_I) and average evenness index $\overline{J(\varepsilon)}$ for *Phillyrea latifolia* in each of the two shrub parts.

Variable	**Upper part**	**Lower part**
D_I	1.65a[1]	1.36b
$\overline{J(\varepsilon)}$	0.67a	0.39b

[1] Means with the same letters within the same variable are not significantly different at the 0.001 level of significance.

Table 6. Interaction means of information fractal dimension (D_I) and average evenness index $\overline{J(\varepsilon)}$ in the upper and lower parts of variously grazed *Phillyrea latifolia* shrubs.

Variable	**Control**		**Grazed treatments**					
			Light		**Moderate**		**Heavy**	
	Upper	**Lower**	**Upper**	**Lower**	**Upper**	**Lower**	**Upper**	**Lower**
D_I	1.61b[1]	1.60b	1.65a	1.28c	1.68a	1.29c	1.66a	1.27c
$\overline{J(\varepsilon)}$	0.63b	0.60c	1.66b	0.34d	0.70a	0.32de	0.70a	0.30e

[1] Means with the same letters within the same variable are not significantly different at the 0.001 level of significance.

Discussion

Phillyrea latifolia

The results produced in this study revealed that *Phillyrea* is developmentally more unstable under grazing than under no grazing (larger leaf fluctuating asymmetry). This is particularly true when we look at both the unreachable and reachable by goats parts of the shrubs together. The separate analyses of the upper and lower parts revealed that *Phillyrea* was able to compensate the stress produced in the lower part by increasing growth and maintaining developmental stability in the upper unrea-

Table 7. Means ± se and F values of the mixed model analysis of covariance for *Cistus monspeliensis*, with shoots nested in individual shrubs and the neperian logarithm of shoot base diameter as covariate. Dependent variables are the scale asymmetry parameters of the relation between internode length and node order (Sa, Sb, R^2adj and $Syx/\overline{y}$), and equation parameters a, b and lnk were derived from the equation $L = kN^{a}e^{-bN}$

Variable	**Grazed treatments**			$\mathbf{F_{2,116}}$	**P**
	Light	**Moderate**	**Heavy**		
Length (shoot)	169.43 ± 33.39	177.97 ± 53.51	175.98 ± 42.27	0.96	0.386
a	0.951±0.523ab[1]	0.835 ± 0.574b	1.105 ± 0.471a	4.92	0.009**
b	-0.260 ± 0.123	-0.213 ± 0.129	-0.242 ± 0.112	2.71	0.071
Sa	0.497 ± 0.165	0.494 ± 0.165	0.452 ± 0.124	1.86	0.159
Sb	0.092 ± 0.045	0.090 ± 0.039	0.079 ± 0.029	2.14	0.122
R^2_{adj}	0.477 ± 0.198	0.416 ± 0.237	0.456 ± 0.201	2.19	0.116
$Syx/\overline{y}$	0.252 ± 0.092	0.256 ± 0.115	0.248 ± 0.110	0.49	0.614
ln*k*	2.264 ± 0.448a	2.184 ± 0.569a	1.935 ± 0.550b	5.87	0.004**

[1] Means with the same letter within the same variable are not significantly different at 0.01 (**) level of significance

chable shoots. It seems that the increased growth of the shoot length of the grazed plants was a reaction to grazing stress in order to counterbalance the removed growth by the browsing animals. Perennial plants usually exhibit a stress tolerance strategy (Grime and Hunt, 1975; Chapin et al., 1993) in their development under dry conditions, i.e. slow growth rate. *Phillyrea*, on the contrary, exhibited a stress avoidance strategy under grazing, i.e. faster growth rate, which is more common in annual plants (Huhta et al., 2000; Vrahnakis, 2000; Alados et al., 2002). It has been shown that grazed and damaged plants produce in many cases more biomass than the control plants (Oesterheld and McNaughton, 1988; Georgiadis et al., 1989). According to Huhta et al., (2000), for compensatory regrowth to take place, the existence of a sufficient amount of undamaged meristems that can lead to branch development is required as well as sufficient resources to support this development. In our case, the tree form of *Phillyrea* allowed enough amount of undamaged shoots and meristems to support the whole plant.

Table 8. Means of information dimension (D_I), and average evenness index $\overline{J_{(\varepsilon)}}$ for Cistus monspeliensis in the three grazing treatments of one-way ANOVA with grazing level as fixed effect factor.

Variable	**Grazed treatments**		
	Light	**Moderate**	**Heavy**
D_I	1.72a[1]	1.64b	1.71a
$\overline{J_{(\varepsilon)}}$	0.73a	0.67b	0.73a

[1] Means with the same letter within the same variable are not significant at the 0.001 level of significance.

The results for *Phillyrea* agree with previous studies. For example, *Anthyllis cytisoides*, also a palatable species, was found to increase its developmental stability under moderate grazing as compared with the light and heavy one (Escos et al., 1997; Alados et al., 1998). The fact that all grazed treatments produced the same response possibly indicates that *Phillyrea* is well adapted to grazing and intensity seems not to affect its development substantially. Probably this reaction is related to the reduction of competition (Belsky, 1987), since it is a dominant and palatable species in the shrublands studied.

The genetic basis of fluctuating asymmetry has been a subject of serious consideration in the literature. Møller and Thornhill (1998) claimed that the presence of fluctuating asymmetry in individuals has a low but significant heritability. In this study, we demonstrated that fluctuating asymmetry can be produced as a result of environmental disturbance without genetic differential influence. The absence of directional asymmetry as well as antisymmetry is not the only proof. The results revealed significant differences of fluctuating asymmetry between the upper and lower parts of the *Phillyrea* shrubs that undoubtedly have the same genotype. The lower part that suffered from grazing showed higher asymmetry values. Moreover, we had no statistically significant differences between the two parts of the shrubs growing in the control area, since both parts were not grazed by animals. In our opinion, this fact suggests that fluctuating asymmetry can be a direct product of developmental noise.

On the other hand, the results of the fluctuating asymmetry analysis are in contrast with the ones produced by the translational asymmetry, since the shrubs of the grazed treatments were found to be less stressed than the control. In fact, the outcome of the two methods is not fully comparable. This is because the translational asymmetry analysis was based on shoots taken only from the upper (above 1.5m height) part of the shrubs, while the latter on leaves taken from both the upper and lower parts of the shrubs. For this reason, the results of the fluctuating asymmetry analysis should be considered as more realistic and a better expression of the grazing effects on *Phillyrea*. On the other hand, the discrepancy of the two methods of measuring developmental instability suggests that for *Phillyrea* it is more important to maintain the structural stability of the shoot than leaf symmetry probably because the production of evergreen leaves is a rather costly physiological process that it is repaid by its increased longevity (Gratani and Bombelli, 1999; Werner et al., 2001).

The results of the fractal analysis of *Phillyrea* branches showed the smaller values of D_I and $\overline{J_{(\varepsilon)}}$ in the grazed treatments, especially at the lower part of the shrubs, which may imply that there was no efficient gas exchange and substances between the plants and the atmosphere (Sugihara and May, 1990; Alados et al., 1999). This lower branch complexity and homogeneity in the grazed plants of *Phillyrea* was paired with their larger developmental instability, found with the fluctuating asymmetry indices, suggesting that grazing is an important stress factor that affects negatively the growth and productivity of this particular species. As a matter of fact, the interaction values of Table 8 suggest that the more *Phillyrea* shrubs are grazed the lower the $\overline{J_{(\varepsilon)}}$ becomes. In this sense, grazing of *Phillyrea* could be considered that leads to productivity deterioration. On the other hand, the fact that grazing, even when it is quite intense, maintains the developmental stability of the top parts of *Phillyrea* shrubs (that are not reached by animals) indicates that this species is very tolerant even under heavy grazing pressure and may suggest that a compromise must be found between overgrazing and non-grazing in pastoral management of this particular species.

Cistus monspeliensis

The results of the translational asymmetry analysis confirm that *Cistus* was not affected by grazing at all including the reduction of competition, especially in the heavily grazed treatment, as it is adapted to stressful areas. This particular species is able to avoid drought stress with a major reduction, almost three-fourth, of its foliage during summer since it is a seasonally dimorphic species (Margaris and Vokou, 1981), and also presents less spatial variability but more temporal changes in leaf angle and leaf area in comparison with other sclerophyllous species (Werner et al., 2001). Thus we can say that it has a rather opportunistic behavior with flexible responses and changes in its structure as an adaptive feature to changing environmental conditions.

The results of fractal analysis also indicate that this species is very well adapted to disturbed situations (e.g. heavy grazing), while under moderate grazing pressure competition with grazing-adapted species may disturb it. Alados et al. (1998) reported that *Chrysothamnus grenii* was also more stable under heavy grazing pressure because it is adapted to stressful areas. These results agree with the ones found by the translational asymmetry analysis which showed a greater shoot elongation in the moderate grazing treatment. Under heavy grazing pressure though, compensatory branching was more important than shoot elongation as the intensity of competition for light with the surrounding species was low (Huhta et al., 2000).

Conclusions

1. The upper, ungrazed part of the palatable shrub *Phillyrea latifolia* maintains its developmental stability when the lower part is grazed, while it becomes unstable when the whole plant is ungrazed indicating that this species is well adapted to grazing pressure. On the other hand, grazing increases leaf fluctuating asymmetry and reduces the fractal dimension of its branches, thus affecting negatively its growth and productivity of this particular species. It seems therefore that a light to moderate grazing is needed to ensure both stability and increased productivity of *Phillyrea* shrubs.
2. Developmental stability of the unpalatable shrub *Cistus monspeliensis* is not affected by grazing, although its branch fractal dimension declines under moderate grazing situations, probably in relation to competition with the other species that are adapted to grazing pressure. Both heavily

disturbed and undisturbed situations favor an increased branch complexity of *Cistus*.

3. Developmental instability and fractal dimension are good indicators of the responses of palatable shrubs to grazing pressure. Both these methods are non destructive to plants, cost effective and produce reliable results. Consequently, they can be used for assessing the effects of grazing management on Mediterranean shrublands.

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References

Alados, C.L., Escos, J. & Emlen, J.M. 1994. Scale asymmetry: a tool to detect developmental instability under the fractal geometry scope. In: Fractals in the Natural and Applied Sciences (Novak, M.M. ed.) pp 25-36. North Holland: Elsevier Sci. B.V. 451pp.

Alados, C.L., Emlen, J.M., Wachochi, B. & Freeman, D.C. 1998. Instability of development and fractal architecture in dryland plants as an index of grazing pressure. Journal of Arid Environments 38:63-76.

Alados, C.L., Navarro, T. & Cabezudo, B. 1999. Tolerance assessment of *Cistus ladanifer* to serpentine soils by developmental stability analysis. Plant Ecology 143:51-66.

Alados, C.L., Giner, M.L, Dehesa, L., Escos, J., Barroso, F.G., Emlen, J.M. & Freeman, D.C. 2002. Developmental instability and fitness in *Periploca laevigata* experiencing grazing disturbance. International Journal of Plant Science 163(6): 969-978 .

Arianoutsou-Faraggitaki, M. 1985. Desertification by overgrazing in Greece: The case of Lesvos island. Journal of Arid Environments 9:237-242.

Belsky, A.J. 1987. The effects of grazing: confounding of ecosystem, community and organism scales. American Naturalist 129:777-783.

Chapin, F.S., Autumn, K. & Pugnaire, F. 1993. Evolution of suites of traits in response to environmental stress. American Naturalist 141 (suppl): S78-S92.

Clarke, G.M. 1993. Patterns of Developmental stability of *Chrysopa perla* L. in response to environmental pollution. Environmental Entomology 22: 1362-1366.

Collins, S.L. 1987. Interaction of disturbances in tall grass prairie: a field experiment. Ecology 68:1243-1250.

Escos, J.M., Alados, C.L. & Emlen, J.M. 1997. Grazing impact on plant fractal architecture and fitness of a Mediterranean shrub (*Anthyllis cytisoides*). Functional Ecology 11:66-78.

Farmer, D., Ott, E. & Yorke, J.A. 1983. The dimension of chaotic attractors. Physica 7D: 153-180.

Freeman, D.C., Graham, J.H.& Emlen, J.M. 1993. Developmental stability in plants: symmetries, stress and epigenesis. Genetica 89: 97-119.

Georgiadis, N.J., Ruess, R.W., Mc Naughton, S.J. & Western, D. 1989. Ecological conditions that determine when grazing stimulates grass production. Oecologia 81: 316-322.

Graham, J.H., Freeman, D.C. & Emlen, J.M. 1993. Developmental stability: A sensitive indicator of population under stress. Environmental Toxicology and Risk Assessment *ASTM STP 1179* (W.G. Landis, J.S. Hughes & M.A. Lewis, eds.) pp. 136-158. American Society for Testing and Materials, Philadelphia.

Gratani, L. & Bombelli, A. 1999. Leaf anatomy, inclination, and gas exchange relationships in evergreen sclerophyllous and drought semideciduous shrub species. Photosynthetica 37(4): 573-585.

Grime J.P. & Hunt, R. 1975. Relative growth-rate: its range and adaptive significance in a local flora. Journal of Ecology 63: 393-422.

Huhta, A. P, Lennartsson, T., Tuomi, J., Rautio, P. & Laine, K. 2000. Tolerance of *Gentiatella campestris* in relation to damage intensity: an interplay between apical dominance and Herbivory. Evolutionary Ecology 14: 373-392.

Lindenmayer, A. 1968. Mathematical models for cellular interaction in development, Parts I and II. Journal of Theoretical Biology 18: 280-315.

Margaris, N. S. & Vokou, D. 1981. Structural and physiological features of woody plants in phryganic ecosystems related to adaptive mechanisms. Ecologia Mediterranea T.VIII: 449-459.

Margaris, N. S. & Koutsidou, E. 1998. Fires and overgrazing: the main driving forces of desertification in the Aegean islands, p. 167-170. In: Ecological Basis for Livestock Grazing in Mediterranean Ecosystems (V.P. Papanastasis & D. Peter, eds). European Commission, EUR 18308 N, Luxemburg.

Markow, T.A. & Clarke, G.M. 1997. Meta- analysis of the heritability of Developmental stability - a giant step backward. Journal of Evolutionary Biology 10:31-37

Martel, J., Lempa, K. & Haukioja, E. 1999. Effects of stress and rapid growth on fluctuating asymmetry and insect damage on birch leaves. Oikos 86:208-216.

Mather, K. 1953. Genetic control of stability in development. Heredity 7:297-336.

McNaughton, S.J. 1977. Diversity and stability of ecological communities: a comment on the role of empiricism in ecology. American Naturalist 111:515-525.

McNaughton, S.J. 1983. Compensatory plant growth as a response to herbivory. Oikos 40:329-336.

Møller, A.P. & Shykoff, J.A. 1999.Morphological developmental stability in plants: patterns and causes. International Journal of Plant Science 160(suppl): S135-S146.

Møller, A.P. & Swaddle, J.P. 1997. Asymmetry, developmental stability and evolution. Oxford University Press, Oxford.

Møller, A.P. & Thornhill, R. 1998. Bilateral symmetry and sexual selection: a meta analysis. American Naturalist 151: 174-192.

Noy-Meir, I. 1998. Effects of grazing on Mediterranean grasslands: the community level. Proceedings of the International Workshop on the Ecological Basis of Livestock Grazing in Mediterranean Ecosystems. (V.P. Papanastasis and D. Peter, eds). October 23-25, Thessaloniki, Greece. pp. 27-37.

Oesterheld, M. & Mc Naughton, S.J. 1988. Intraspecific variation in the response of *Themela triandra* to defoliation: the effect of time of recovery and growth rates on compensatory growth. Oecologia 77: 181-186.

Palmer, A.R. 1994. Fluctuating asymmetry analyses: a primer. In: Developmental instability: its origins and evolutionary implications (T.A. Markow, ed.), pp. 335-364. Kluwer, Dordrecht, Netherlands.

Palmer, A.R. & Strobeck, C. 1986. Fluctuating asymmetry: measurement, analysis, patterns. Annual Review of Ecology and Systematics 17:391-421.

Palmer, A.R. & Strobeck, C. 1992. Fluctuating asymmetry as a measure of developmental stability: implications of non-normal distributions and power of statistical tests. Acta Zoologica Fennica 191: 57-72.

Papanastasis, V.S., Kyriakakis, S. & Ispikoudis, J. 1990. Forestry and grazing practices in Cretan Landscape. pp. 42-46. Petromarula 1. Corpus Christi College, Cambridge, England.

Perfectti, F. & Camacho, J.P.M. 1999. Analysis of genotypic differences in developmental stability in *Annona cherimola*. Evolution 53: 1396-1405.

SAS Institute, 1995. SAS/STAT user’s guide, version 6. SAS institute, Cary, N. C.

Shapiro, S.S., Wilk, M.B. & Chen, H.J. 1968. A comparative study of various tests for normality. Journal of American Statistical Association 63: 1342-1372.

Smith, T.G., Lange, G.D. & Marks, W.B. 1996. Fractal methods and results in cellular morphology - dimensions, lacunarity and multifractals. Journal of Neuroscience Methods 69: 123-136.

Sokal, R.R. & Rohlf, F.J. 1995. Biometry. W.H. Freeman, New York.

Sugihara, G. & May, R.M. 1990. Applications of fractals in ecology. Trends in Ecology and Evolution 5: 79-86.

Swaddle, J.P., Witter, M.S. & Cuthill, I.C. 1994. The analysis of fluctuating asymmetry. Animal Behavior 48: 986-989.

Vrahnakis, M.S. 2000. Mathematical Models in the Study of Range Plant Species Dynamics. PhD thesis. Aristotle University of Thessaloniki, Greece, pp. 176 (In Greek with English summary).

Waddington, C.H. 1957. The Strategy of Genes. Allen and Unwin, London.

Werner, C., Ryel, R.J., Correia, O. & Beyschlag, W. 2001. Structural and functional variability within the canopy and its relevance for carbon gain and stress avoidance. Acta Oecologica 22: 129-138.

Zar, J.H. 1984. Biostatistical Analysis. Prentice Hall, Englewood Cliffs, New Jersey.