



# Contrasting patterns in leaf traits of Mediterranean shrub communities along an elevation gradient: measurements matter

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**Abstract** We assessed the changes in community-weighted mean (CWM) and variability of specific leaf area (SLA) and leaf area (LA) of different Mediterranean shrub communities along an elevation gradient in the island of Sardinia (Italy). Furthermore, we explored the relative contribution of species turnover and intraspecific variation to shifts in CWM values along the gradient. Forty sampling units (5 × 5 m) were selected in a probabilistic way along a 1300 m elevation gradient which crossed four thermotypes

(thermometric belts). Leaf traits were measured in each sampling unit. ANOVA and a trend test for monotonic changes in variance were used to assess, respectively, CWM differences and variability in both the leaf traits across thermotypes. Variance decomposition of CWM values was used to identify the role of inter- and intraspecific variation. SLA and LA responded differently along the studied gradient in terms of abundance-weighted mean values and variability: CWM of SLA showed the lowest values in the driest thermotype, while LA in the more humid one; SLA variability showed a significant increasing trend with increased water availability, while LA variability did not show any pattern. The contribution of intraspecific trait variation was significant for both the leaf traits, but higher for SLA, where negative

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covariation between inter- and intraspecific variation was detected. We highlight the importance of simultaneously considering measurements of both leaf traits to understand the functional response of communities in Mediterranean environments. Moreover, neglecting intraspecific variation in leaf traits, even along steep gradients with relevant species compositional changes, can result in the underestimation of the amount of trait variation in response to environmental changes.

**Keywords** Community weighted mean · Interspecific and intraspecific variation · Leaf area · Plant functional traits · Specific leaf area · trait–environment relationship

## Introduction

A large number of studies have identified relationships between traits and environmental conditions (Garnier et al. 2016 and references therein), such as climate and soil properties at different scales and in different regions and systems (e.g. Ackerly 2004; Wright et al. 2004; Ordóñez et al. 2009). However, the extent to which such patterns can be generalized beyond the studied scale/region/system is still unknown (Shipley et al. 2016). In particular, despite the Mediterranean biogeographic region being one of the world's most important reservoirs of plant diversity (Medail and Quezel 1999) and one of the most vulnerable areas of the planet to climate change (Thuiller et al. 2005; Cuttelod et al. 2009; Moreno and Oechel 2012), the trait–environment relationship is still poorly investigated (Chelli et al. 2019). Here, temperature is expected to increase (Senatore et al. 2011) together with an increase in weather extremes in both the sense of heavy rainfall and summer heat waves, with a high incidence of drought (ISAC-CNR 2009). In this context, shedding light on the trait–environment relationship along temperature and water stress gradients can help to interpret vegetation response under changing climatic conditions (Scherrer et al. 2017).

In the Mediterranean biogeographic region, shrublands are one of the most important and widespread vegetation types (Naveh and Whittaker 1979; Cowling et al. 1996; Shoshany and Karnibad 2011). Such vegetation is dominated by evergreen shrubs usually

with small and sclerophyllous leaves as part of their drought tolerance strategy (Mooney and Dunn 1970).

Of all plant traits, those of leaves are among the most important when it comes to water-use strategies (Wilson et al. 1999; Vendramini et al. 2002). Specific leaf area (SLA, the light-capturing surface area per unit of dry biomass, Poorter et al. 2009), is generally recognized as one of the most significant trait to study water and nutrient availability impacts on plant growth (Wright et al. 2001; Reich 2014; Wellstein et al. 2017). Leaf area (LA) is another trait which has significant implications regarding the regulation of leaf temperature and water-use efficiency during photosynthesis (Ackerly et al. 2002). SLA and LA are two fundamental traits influencing community structure and functioning of Mediterranean ecosystems (Gross et al. 2013).

In community level studies based on average trait values for species, LA and SLA usually showed parallel changes, which suggests that these traits are linked in their functional strategy associated with water and/or nutrient availability (Ackerly et al. 2002 and references therein). Accordingly, global studies showed a decrease in LA and SLA along with decreasing water and/or nutrient availability (Poorter et al. 2009; Wright et al. 2017). However, comparative studies accounting for species trait variability suggested that LA and SLA may not be strictly related (e.g. Ackerly et al. 2002), thus highlighting the importance of approaches taking into account both leaf traits and their intraspecific variation along environmental gradients.

Decreased plant growth rates are associated with lower SLA values (Wellstein et al. 2017 and references therein), and are often found under conditions of water stress (Chapin 1991), as they can improve water-use efficiency (Wright et al. 2001). LA shows a similar trend, with small leaves reducing resistance in the boundary layers and allowing the plant to maintain lowered temperatures and higher photosynthetic water-use efficiency when solar radiation is high and water availability is low (Wright et al. 2001; Ackerly et al. 2002). In short, SLA and LA are good proxies of broad resource-use strategies and are negatively correlated with many other hard and soft leaf traits (e.g., leaf toughness, leaf longevity, leaf dry matter content, stomatal conductance; Wright et al. 2002; Reich et al. 2003; Wright et al. 2004; Hoffmann et al. 2005).

Few papers deal with changes in leaf traits along environmental gradients of shrubs communities in Mediterranean-type climates (e.g., Ackerly et al. 2002; Ackerly 2004; Anacker et al. 2011, conducted in North American chaparral), confirming the above described patterns of SLA and LA. Additional studies used leaf traits to explore assembly rules along aridity gradients in shrublands of the Mediterranean basin (e.g., Gross et al. 2013; Le Bagousse-Pinguet et al. 2015).

Looking at a single trait allows the range and distribution of values to inform the relationship between ecosystem function and community structure (Ackerly et al. 2002). To summarize the functional composition of plant communities for a certain trait taking into account species abundances, Garnier et al. (2004) proposed the so-called ‘community-weighted mean trait values’ (CWMs).

Changes in community composition along climate and soil gradients are expected to be strongly represented in leaf traits, due to their reflection of water-use strategies (Le Bagousse-Pinguet et al. 2017). Increased variability in these leaf traits is expected when conditions are favorable for plant life, e.g., water and temperature stress are reduced; soils are more fertile (Ackerly et al. 2002; Hoffmann et al. 2005; Cornwell and Ackerly 2009). In contrast, habitat filtering may occur in the face of extreme aridity or a reduction in water availability, reducing the distribution of SLA and LA values within the community, and suggesting that species with high stress tolerance do not generally inhabit broad environmental ranges (Thuiller et al. 2004).

Most studies focusing on trait–environment relationships along gradients used only mean trait values per species (usually weighted by species proportions), assuming that between-species trait variation is generally considerably greater (e.g. Cornelissen et al. 2003) than within-species variation. However, it is clear that some traits are more variable than others (Siefert et al. 2015), and a growing number of studies confirm that intraspecific variation could play a relevant role to shifts in community-level trait measures (i.e., Lepš et al. 2011; Kichenin et al. 2013) and assembly rules (i.e., Jung et al. 2010; Le Bagousse-Pinguet et al. 2015) along environmental gradients. In particular, both SLA (Wellstein et al. 2013, 2017; Liancourt et al. 2015) and LA (Fraser et al. 2009) showed a certain intraspecific variation, with SLA

demonstrating a larger one (Rozendaal et al. 2006; Siefert et al. 2015).

In this paper, we aimed to assess the patterns of community-level leaf traits (namely, SLA and LA) along an elevation gradient (1300 m) spanning four thermotypes or thermometric belts, defined on the basis of temperature data condensed in two indexes (It = thermometric index; Tp = positive annual temperature), following Rivas-Martínez et al. (2011) method, and mapped by Canu et al. (2015) in Mediterranean shrubland ecosystems in the island of Sardinia (Italy). We measured leaf traits at plot level to take into account intraspecific variability.

In particular, we propose the following hypotheses:

H1 SLA and LA at community level should be significantly lower in the most arid climate (thermotype 1) compared to the most cool and moist environment (thermotype 4).

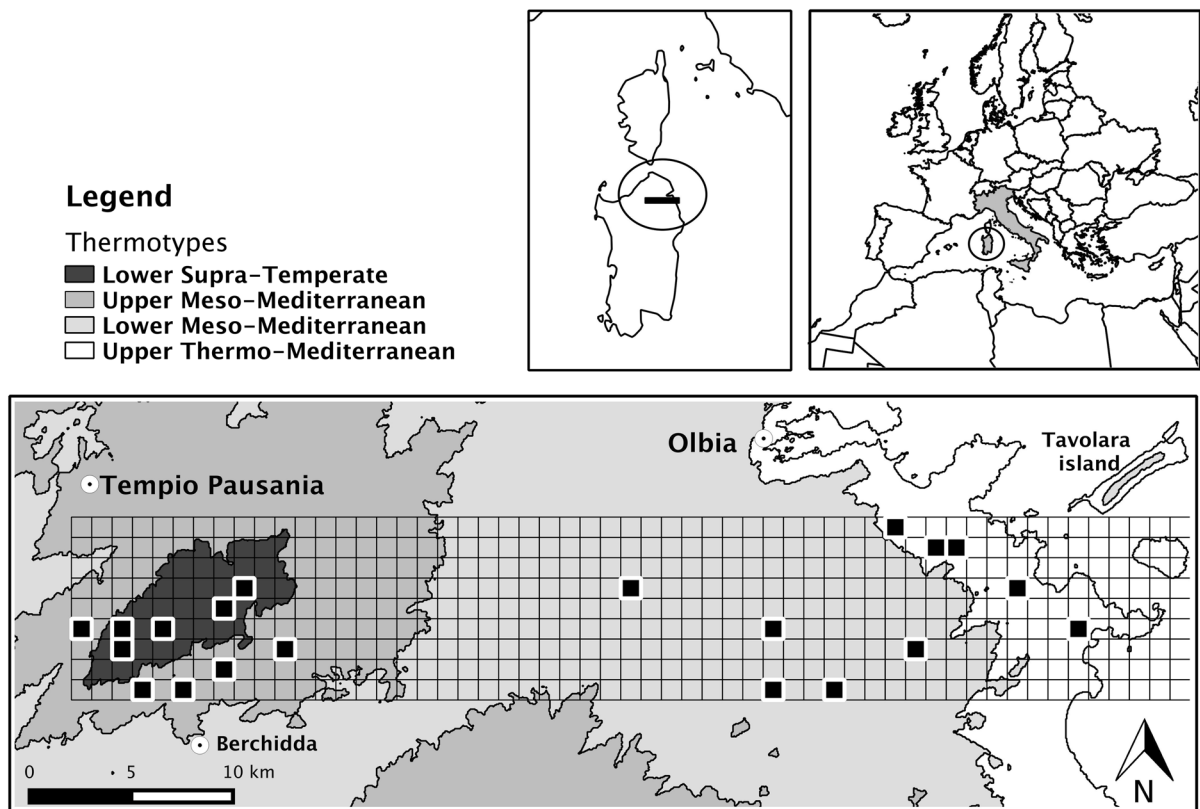
H2 The variability of both leaf traits (in terms of variance) should increase (niche differentiation) toward the cooler and moister end of the gradient (thermotype 4).

Furthermore, we disentangled the contribution of interspecific (i.e. species turnover) and intraspecific trait variation to shifts in CWM values along the gradient, hypothesizing (H3) a significant influence of intraspecific variation in both leaf traits.

## Materials and methods

### Study area

Sardinia, the second largest island in the Mediterranean Sea (ca. 24,090 km<sup>2</sup>), has a unique flora as a result of the unique geological history that played an essential role in shaping plant diversity (Medail and Quezel 1999; Mansion et al. 2008). The Sardinian flora consists of 2301 native vascular plants (Bartolucci et al. 2018), of which more than 170 are endemic to the island: concentrated mainly in the mountains (Cañadas et al. 2014). This study has been conducted in the NE part of Sardinia (Fig. 1), locally known as the Gallura subregion, with constant geology in the altitudinal gradient, being characterized by Hercynian granites, having homogeneous slopes and patterns of degradation. Furthermore, the selected area



**Fig. 1** Study area and scheme of the sampling design. T1–T4 gradient is represented by white to dark grey areas. The selected sampling quadrats are highlighted in black; each quadrat includes two sampling units  $5 \times 5$  m

is also considered homogeneous from a biogeographic standpoint (Fenu et al. 2014). Altitude range in this area is from 0 (coast) to 1354 m (top of Mt. Limbara massif). This altitudinal gradient allows five different thermotypes to occur in the study area (Canu et al. 2015). Thermotypes are described on the basis of thermicity indexes developed by the Worldwide Bioclimatic Classification System (Rivas-Martínez et al. 2011). Annual mean temperature ranged from 16.6 °C in the coastal area to 10.5 °C at the top of Mt. Limbara; annual rainfall ranged from 560 to 1160 mm.

Potential natural vegetation of the area (sensu Farris et al. 2010) is represented by different forest types depending on the thermotypes: the coastal Thermo-mediterranean belt is characterized by the presence of Phoenician juniper vegetation (*Oleo-Juniperetum turbinatae* and *Erico-Juniperetum turbinatae* on southern and northern slopes, respectively). Wild olive (*Olea europaea*) vegetation is prevalent in the lower Mesomditerranean belt, holm-oak (*Quercus*

*ilex*) vegetation prevails at both the upper Mesomediterranean (*Galio-Quercetum ilicis*) and sub-Mediterranean and Temperate belts (*Saniculo-Quercetum ilicis*).

The intense past sylvo-pastoral management, that included the use of fire and extensive plantations of alien trees, changed the landscape and determined the current situation where the shrublands represent the most abundant plant communities present in the area.

#### Sampling design

To study a strong climatic gradient, we needed strata that could represent the range of temperatures and water stresses characterizing our study area, stretching from the coastline to the inner areas of Sardinia. To describe this gradient, we adopted a modified version of the thermotypes recognized in the bioclimatic map of Sardinia (Canu et al. 2015) where we unified the upper belts into a unique “temperate” cacuminal belt, to obtain four different thermotypes describing the

whole transect (Fig. 1): upper Thermomediterranean (T1); lower Mesomediterranean (T2); upper Mesomediterranean (T3) and lower Supratemperate (T4).

The study area was defined creating a grid of 1 km<sup>2</sup> units including the Limbara massif and extending eastward toward the Olbia coastline (40 km gradient): regarding the other relevant environmental factors, we can consider geology (also soil), erosion patterns, history (including fire regime and cutting) and animal influence as constant along the bioclimatic gradient.

From the original 495 quadrats, we excluded (i) all the mixed quadrats crossing two thermotypes, and (ii) all quadrats showing a land cover characterized by < 50% of shrubs and garrigues (CLC Regione Sardegna). Among the remaining quadrats, within each of the four strata we selected five sampling quadrats; in each sampling quadrat, we performed a random selection of two sampling units 5 × 5 m. As a whole, we sampled 40 sample units of 25 m<sup>2</sup> divided in four thermotypes (two sample units × 5 quadrats × 4 thermotypes) where we registered the complete species list and visually estimated the species coverage (%) during May 2016 (Online Appendix 1).

In each sampling unit, we selected all the species contributing to reach a relative cumulative coverage of 80%. For each species, five leaves in full sun were sampled from five mature and healthy individuals; such sampling size adequately captures leaf traits intraspecific variability in Mediterranean contexts (Petruzzellis et al. 2017). In total, 900 individuals belonging to 33 species, including shrubs and herbs, were selected for leaf traits measurements (Online Appendix 1). We measured SLA and LA following standard protocols (Pérez-Harguindeguy et al. 2013): leaf area was measured within few hours after collection; subsequently leaves were oven-dried for 72 h at 70 °C and leaf dry mass was measured to calculate SLA. Community weighted mean values (CWM, Garnier et al. 2004) were then calculated according to species plot-level cover values (i.e., we averaged the species trait values from individual measurements taken in a given plot and used it to calculate CWM of that plot). According to recent methodological studies (e.g. Peres-Neto et al. 2017; Zelený et al. 2018), measuring traits for each species at each site, and using these plot-level trait measurements to calculate CWM, avoid the classic problem with inflated Type I error.

## Data analysis

We analyzed the data to test the differences existing along the gradient, according to the following variables: (i) community composition—to observe differences in terms of species composition and abundance we performed a NMDS (standardized samples by total, Bray–Curtis resemblance). We integrated NMDS results with PERMANOVA (Anderson 2001; fixed factor: thermotypes, 9999 permutation, unrestricted permutation of raw data, Bray–Curtis resemblance) and a PERMDISP analyses, a test of the homogeneity of multivariate dispersions within groups, as PERMANOVA makes the implicit assumption that dispersions are roughly constant across groups; (ii) changes of CWM values—to test if CWM values of SLA and LA significantly changed according to the four thermotypes (H1), we used ANOVA for multiple samples and Bonferroni post hoc test; (iii) trend in leaf traits variance—to test for changes in the variance of leaf traits along the gradient (H2), we used a trend test for monotonic trend in variance suggested by Neuhauser and Hothorn (2000). The test is based on the finite-intersection approach, the Brown–Forsythe transformation, and Kendall’s tau coefficient (Noguchi and Gel 2010; Gastwirth et al. 2015). The finite-intersection approach (Mudholkar et al. 1993) combines p values of the component test statistics, which correspond to a finite number of nested hypotheses. Fisher’s p value combination method (Fisher 1934) is used in this study. Finally, to distinguish (iv) the relative contribution of inter- and intraspecific variation on community-level trait values (H3) we followed the approach proposed by Lepš et al. (2011). This method is based on the decomposition of the total sum of squares ( $SS_{\text{specific}}$ ) of the plot-level trait variance related to a certain environmental variable into “interspecific” (or “fixed”) ( $SS_{\text{interspecific}}$ ), “intraspecific” ( $SS_{\text{intraspecific}}$ ) and “covariation” ( $SS_{\text{cov}}$ ) components, so that  $SS_{\text{specific}} = SS_{\text{interspecific}} + SS_{\text{intraspecific}} + SS_{\text{cov}}$  (Kichenin et al. 2013). First, for each plot, we calculated “specific” plot-average trait values using species trait values as measured on that plot (which includes both inter- and intraspecific components), and “interspecific” (or “fixed”) plot-average trait values using species trait values averaged over all plots along the selected environmental gradient (which takes into account only species turnover and removes the

intraspecific variability component). Second, we calculated “intraspecific” plot averages as the difference between “specific” and “interspecific” plot-average trait values (thus removing the component of interspecific variability). Thirdly, the “covariation” component (i.e., the effect of covariation between interspecific and intraspecific trait variation) was obtained as follows:  $SS_{cov} = SS_{specific} - SS_{interspecific} - SS_{intraspecific}$ . As environmental variables we used both categorical (i.e., thermotype) and continuous ones (i.e., temperature seasonality and Potential EvapoTranspiration, PET; Canu et al. 2015).

All statistical analyses were performed in R, version 3.2.2 (R Development Core Team 2015). In particular the following R packages were used: package *vegan* (function *betadisper*) for the analysis of multivariate homogeneity of group dispersion; package *lawstat* (function *neuhausser.hothorn.test*) for the trend test for monotonic trend in variance (Neuhausser and Hothorn 2000); Package *cati* (function *traitflex.anova*) for the analysis on the relative contribution of inter- and intraspecific variation on community-weighted mean values (Lepš et al. 2011).

## Results

### Community composition and species assemblages

We found shrub communities, physiognomically referred to as *Euphorbio dendroidis-Calicotometum villosae* prevalent at the upper Thermomediterranean belt (T1), *Pistacio lentisci-Calicotometum villosae* dominant at the lower Mesomediterranean (T2), *Erico-Arbutetum* widespread at the upper Mesomediterranean (T3) and *Ericetum scopario-arboreae* dominating at sub-Mediterranean and Temperate belts (T4, Farris et al. 2007; Galié et al. 2015). Plant communities differ between thermotypes (PERMANOVA  $p < 0.05$  for all pairwise tests; PERMDISP  $p > 0.05$ ). The sub-Mediterranean and Temperate *Ericetum scopario-arboreae* (belt T4) is the community that mostly diverged from the others (NMDS, Fig. 2), on the contrary *Euphorbio dendroidis-Calicotometum villosae* of the Thermomediterranean belt (T1) is the community that, being statistically different from the others, appeared less distinct from *Pistacio lentisci-Calicotometum villosae* and *Erico-Arbutetum* (T2 and T3).

### Community-level trait variation

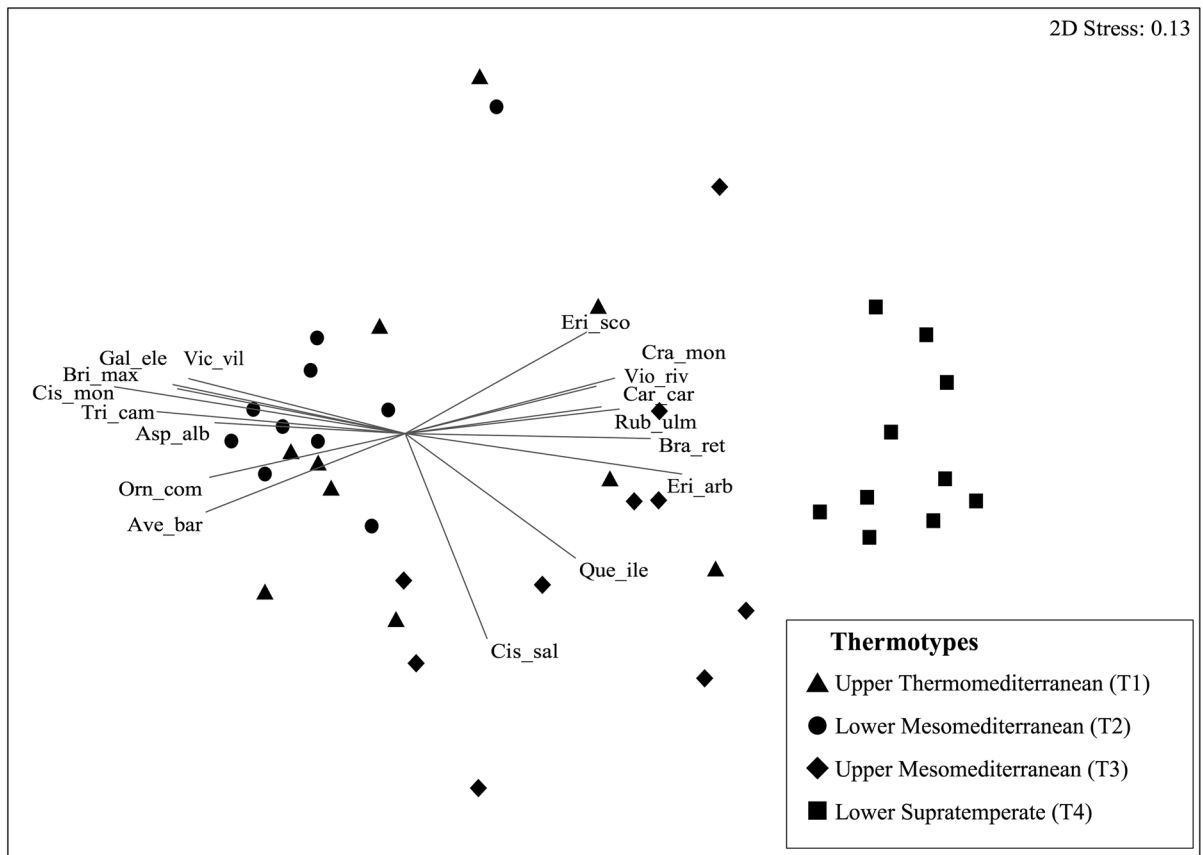
Significant differences between community-level SLA values have been found ( $p < 0.05$ ). In detail, T1 (SLA =  $8.83 \text{ mm}^2 \times \text{mg}^{-1}$ ) and T4 (SLA =  $13.51 \text{ mm}^2 \times \text{mg}^{-1}$ ) were significantly different according to Bonferroni post hoc test ( $p < 0.05$ ; Fig. 3a). Furthermore, we observed a significant increase in variability along the gradient from T1 to T4 (Test Statistic = 3.115;  $p < 0.01$ ). Regarding LA, ANOVA showed significant differences between thermotypes ( $p < 0.05$ ). Differences were found between T2 (LA =  $410.04 \text{ mm}^2$ ), T3 (LA =  $407.87 \text{ mm}^2$ ) and T4 (LA =  $102.33 \text{ mm}^2$ ;  $p < 0.05$ ; Fig. 3b), with the lowest mean values found at T4. No significant trends in LA variability were detected along the gradient.

### Contribution of inter- and intraspecific variation

The contributions of inter- and intraspecific variability in explaining the response of community-level SLA and LA variation to the three environmental variables (i.e., thermotype, temperature seasonality, PET) were both significant only for “thermotypes” ( $p < 0.01$ ; Fig. 4). On the contrary, along the temperature seasonality gradient the contribution of species turnover was significant only for SLA ( $p < 0.05$ ), while along the PET gradient both the components were not significant for both leaf traits. For SLA, the total variability effects were significant for all the three variables ( $p < 0.05$ ) and was highest for the variable “thermotype” (19.61%). Here, species turnover and intraspecific variability showed a similar contribution (turnover = 42.05%; intraspecific = 36.04%), but there was a strong negative covariation (− 58.48%) between the effects of inter- and intraspecific variability on CWM values of SLA. For LA, the total variability effects were significant only for “thermotype” (26.36%;  $p < 0.01$ ). Here species turnover showed a higher contribution (23.88%) with respect to intraspecific variability (12.25%), with a negative covariation (− 9.77%).

## Discussion

Our study explored the leaf trait-environment relationship along an elevation gradient across four



**Fig. 2** Non-metric multidimensional scaling (NMDS) plot of a two-dimensional solution (Bray–Curtis similarities, standardized samples by total abundance data—stress 0.13). We represented the species showing a higher correlation to the ordination axes (Spearman  $\rho > 0.6$ ). Species are labeled as follows: Ave\_bar: *Avena barbata*; Orn\_com: *Ornithopus compressus*; Asp\_alb: *Asparagus albus*; Tri\_cam: *Trifolium*

thermotypes in Mediterranean shrubland ecosystems. The large environmental gradient included four types of compositionally different plant communities. SLA and LA showed different patterns in both abundance-weighted values and variability along the gradient. Additionally, the studied leaf traits differed in the relative contribution of species turnover and intraspecific variation to their changes at community level along the gradient, with SLA showing a relevant contribution of intraspecific variation, higher with respect to LA. Overall, our results highlight the importance of approaches considering simultaneously different traits and their intraspecific variability, even in studies encompassing steep environmental

gradients (Lepš et al. 2011; Kichenin et al. 2013; Derroire et al. 2018; Garnier et al. 2018).

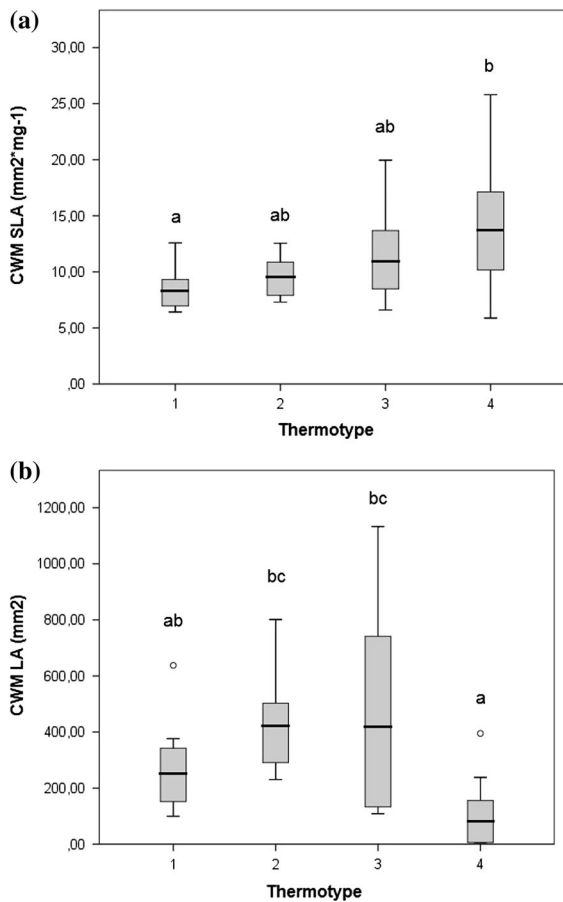
Changes in CWM values and variability

Our results showed significantly different values of community-level SLA between the two extremes of the gradient, with lower values in the driest thermotype. On the contrary, community-level LA values were significantly different only between T2 and T3 and T4, leading us to confirm H1 of lower leaf traits values in the most arid thermotype only for SLA. Regarding trait variability, our hypothesis (H2) of increasing variability of both leaf traits toward the cooler and moister end of the gradient (T4) is

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**Fig. 3** Boxplots of community-level SLA (a) and LA (b) for each thermotype (increasing elevation and decreasing heat and water stress from T1 to T4). Significant differences between mean values are indicated by different letters according to ANOVA ( $p < 0.05$ )

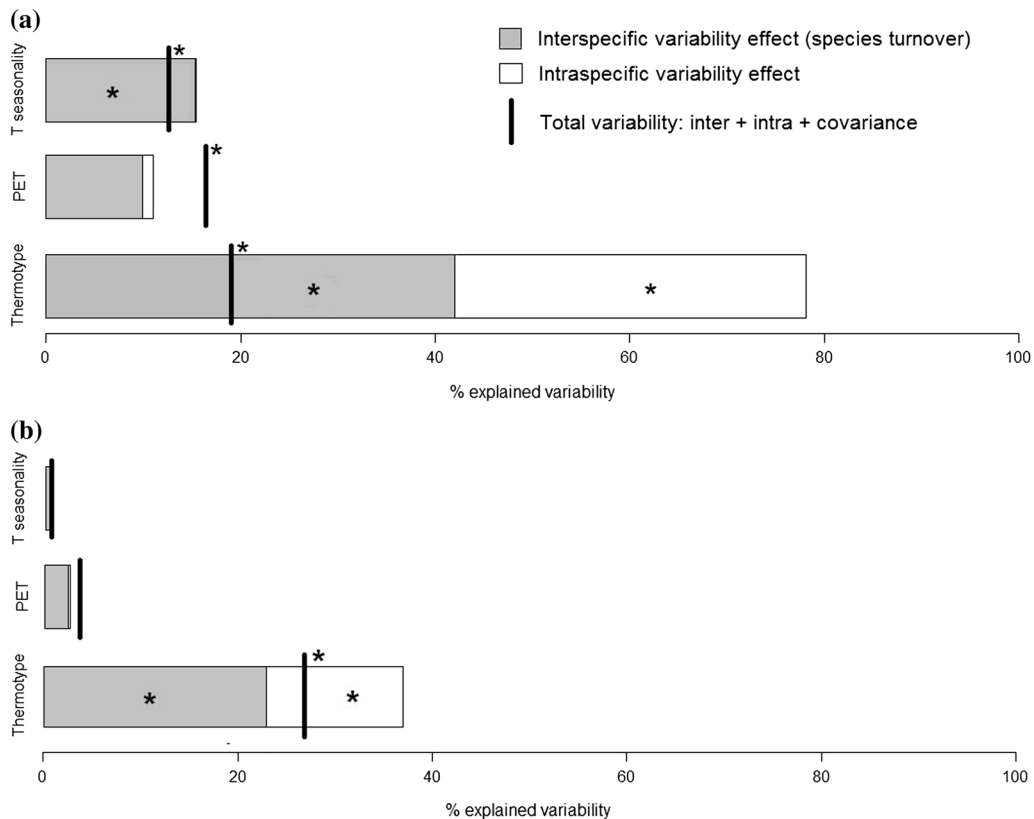
confirmed only for SLA, while LA showed the lowest level of variability in the cooler and moister extreme of the gradient. Variation in the patterns of SLA and LA in the present study across the climatic gradient confirm the expectation that the two leaf traits are not strictly correlated if species variability is properly accounted. The advantage of looking at variation within a trait (instead of using average and fixed trait values) allows us to separate the behavior of the two traits (as in Ackerly et al. 2002).

The lowest community-level SLA values were found in the driest thermotype, which reflects the increase in water-use efficiency in a more arid environment (Wright et al. 2001; Wellstein et al. 2017). Low SLA values and decreased plant growth rates usually reflect water stress (Chapin 1991).

Moreover, T1 is characterized by the lowest SLA variability, most likely indicating a strong habitat filtering generating a convergence in leaf strategy to cope with drought in our system (see also Wright et al. 2002). Under drought conditions, leaves tend to have a denser mesophyll layer, and cells with thicker cell walls, this helps avoid wilt (Shield 1950; Poorter et al. 2009; Wellstein et al. 2017). The increase in SLA variability along the gradient can be explained by the occurrence of more benign conditions, thus allowing the coexistence of species/individuals with different functional strategies related to water-use based on different mechanisms of coexistence (see Wellstein et al. 2014).

Low LA generally leads to high water stress tolerances, whereas species equipped with large leaves are better suited to mesic environments (Westoby et al. 2002). Accordingly, we would expect larger and more variable LA due to more benign conditions in terms of temperature and water availability in T4. However, our results indicated that different habitat filtering mechanisms are acting on this trait leading to both low values of abundance-weighted mean and variability in the two extremes of the gradient. Our findings are consistent with those of Gross et al. (2013), who found a quadratic relationship between functional diversity and mean LA values along a rainfall gradient in Mediterranean shrublands. We propose different explanations for the unexpected strong convergence to low mean LA values in T4. First, LA is a key trait that is strongly influenced by air temperature (Wright et al. 2017). Nocturnal low temperatures in particular seem to have the most significant impact on LA (Wright et al. 2017 and references therein). The T4 is in fact the only thermotype having average lower temperatures in the coldest month approaching 0 °C (0.4 °C in the locality Vallicciola at 1040 m a.s.l. on Limbara massif), and an average snow cover of 3–4 weeks every year (Farris et al. 2007). Second, functional trade-offs between different plant traits could be significant in combined trait responses to increase the variety of ways that plants can respond to environmental stress (Givnish 1984; Marks and Lechowicz 2006; West et al. 2012). Thirdly, LA is more strongly anchored to species identity than SLA, and thus more responsive to species turnover. These results correspond to the ones of Kichenin et al. (2013) who found a major contribution of interspecific variability in explaining the response of LA to





**Fig. 4** Decomposition of total variability in community weighted mean SLA (**a**) and LA (**b**) values into interspecific (species turnover), intraspecific and covariation effect explained by temperature seasonality, PET and thermotypes. Covariation strength is represented by the interval between the “total variability” and the sum of inter- and intraspecific variability effects. A value of total variability that is lower than the sum of

inter- and intraspecific variability effects indicates negative covariations, and a value of total variability that is higher than the sum of inter- and intraspecific variability effects indicates positive covariations. Statistical significance (\*) of inter- and intraspecific and total variability effects are indicated on the graph, when significant

elevation. Since T4 is characterized by plant communities that mostly diverged from the others in terms of species composition, here the compositional changes were reflected by dramatic changes in functional strategy in terms of LA. Accordingly, plant communities of T4 were dominated by *Erica arborea* and *Erica scoparia*, both characterized by small persistent linear leaves. On the contrary, for SLA the species compositional changes between T2-3 and T4 were not reflected by a functional shift. In other words, the different plastic behavior of these two leaf traits could be behind the unexpected pattern of LA in T4 outlining the relevance of intraspecific variation. In contrast to the majority of many other leaf traits, SLA is related to the cell volume ratio of leaf parenchyma to epidermis cells, making it more plastic than traits dependent on

subcellular processes (Kichenin et al. 2013). Mitchell and Bakker (2014) found SLA variation to be mostly based on plasticity with low relevance of ontogeny or local adaptation. For size dependency of SLA and further discussion see Milla et al. (2008).

#### The relative contribution of intraspecific variation

Our finding of a similar relative contribution of inter- and intraspecific community-level SLA variation (55% and 45%, respectively) along the gradient of four thermotypes, confirms the plasticity of this trait (Liancourt et al. 2015; Siefert et al. 2015; Wellstein et al. 2017). Kichenin et al. (2013) found similar patterns along a 900 m elevation gradient spanning subalpine and alpine plant communities in New

Zealand. However, the effect of species turnover can be more marked along other environmental gradients (here, e.g. with temperature seasonality), as also showed by Lepš et al. (2011). Regarding LA, the relative contribution of species turnover is twice as big as intraspecific variation (66.1% and 33.9%, respectively) along the gradient of four thermotypes. Despite the plastic behavior that this trait has shown (Fraser et al. 2009), we demonstrated that its changes at the community level were mostly dependent on species turnover (Rozendaal et al. 2006; Siefert et al. 2015). These results lead us to confirm H3 on a significant influence of intraspecific variation in both leaf traits, but with a larger role of this component for SLA. It is most likely that SLA varies with high-temperature and low-water availability at the intraspecific level (Ackerly 2004; Poorter et al. 2009; Wellstein et al. 2017) which are known to strongly decrease with increasing elevation in Mediterranean contexts.

Moreover, our results provide relevant implications for future studies focusing on leaf trait–environment relationship in Mediterranean contexts. Indeed, the measurements of individual traits within species show that trait-based community responses to environmental changes are not adequately predicted using leaf traits mean values (Lepš et al. 2011). This is in part due to the influence of intraspecific trait variation (Garnier et al. 2018; Kichenin et al. 2013). In particular, the significantly negative covariation between inter- and intraspecific SLA variation suggests that there is a negative compensation between them. For example, in species with high SLA, individuals with lower SLA values may be promoted, and in species of lower SLA, individuals with higher values are promoted. This topic is worthy of further research, as significant uncertainty remains about the complex responses of plant communities to the environment (Lepš et al. 2011).

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#### Compliance with ethical standards

**Conflict of interest** The authors declare that they have no conflict of interest.

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