

# Plant size and local abiotic factors determine the intra-specific variation in the multi-stemmed architecture of *Prunus lusitanica* at the Northeast limit of its global distribution

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## Abstract

**Aim of study:** The present work provides novel insights on factors (either intrinsic or extrinsic) that trigger sprouting in woody species living at range margins. We aim to explain the inter-individual variability in the multi-stemmed architecture of *Prunus lusitanica* L., an Iberian evergreen relict tree related to the Tertiary flora.

**Area of study:** Northeastern Mediterranean mountains of the Iberian Peninsula, the Northeast limit of the global distribution of the species.

**Material and methods:** We gathered data on two modes of vegetative reproduction, basal and layering sprouts, in 288 clumps of *Prunus lusitanica* from four populations. We modeled and analyzed the effect of environmental factors (topography, canopy cover, soil moisture and disturbances) and plant size (diameter at breast height) on sprouting by means of Generalized Linear Model and other statistical approaches.

**Main results:** Plant size arises as the principal factor to explain the variability of the numbers of both types of sprouts yet it is not a trigger factor. Natural and anthropogenic disturbances promote basal and layering shoots, while tree canopy is mainly relevant for basal shoots, and slope and soil moisture are significant factors for layering shoots.

**Research highlights:** The multi-stemmed architecture of *P. lusitanica* at the Northeastern limit of its worldwide distribution is triggered by local environmental factors and disturbances. Each external factor shows different levels of influence on the variability and type of vegetative reproduction yet the intensity of the response is driven by the size of the largest trunk of each clump.

**Key words:** vegetative reproduction; sprouting; disturbances; woody plants; relict tree; subtropical; Iberian Peninsula.

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## Introduction

Vegetative reproduction is a primitive character originated independently in numerous lineages throughout the evolution of angiosperms (Klimês *et al.*, 1997) and is currently the dominant reproduction mode in a wide range of communities in different biomes and geographical regions (Klimês *et al.*, 1997; Peterson and Jones, 1997; Fernández-Palacios and Arévalo, 1998; Bond and Midgley, 2001, 2003). It involves a wide variety of mechanisms (Harper, 1977; Klimês *et al.*, 1997; Del Tredici, 2001) providing several benefits such as indefinite persistence of individuals and populations (Bond and Midgley, 2001;

Tanentzap *et al.*, 2012) and the efficient gathering of resources due to the physiological integration of potential resulting clones (Jonsdottir and Watson, 1997; Klimês *et al.*, 1997).

The multi-stemmed architecture of some woody species arises from sprouting which is a common type of vegetative reproduction (Bond and van Wilgen, 1996; Peterson and Jones, 1997; Del Tredici, 2001; Bellingham and Sparrow, 2009; Tanentzap *et al.*, 2012). It is considered an adaptive advantage for the persistence of populations (Jonsdottir and Watson, 1997; Peterson and Jones, 1997; Bond and Midgley, 2001) since it reduces dependence on sexual reproduction in stressful environments (Bond and Midgley, 2001) and improves the resilience of individuals after disturbances (Bellingham *et al.*, 1994; Bond and Van Wilgen, 1996; Peterson and Jones, 1997;

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Kammesheidt, 1999; Bond and Midgley, 2003; Bellingham and Sparrow, 2009). Sprouting is a successful response against poor light environments (Peterson and Jones, 1997; Tanentzap *et al.*, 2012) or a lack of nutrients (Bellingham and Sparrow, 2009) and is also a common response in plants after logging, fire, herbivory or wind storms (Bellingham *et al.*, 1994; Bond and Van Wilgen, 1996; Kammesheidt, 1999; Bond and Midgley, 2003). However disturbances are not always responsible for multi-stemmed architecture development. Sprouting also varies along species' lifespan and with plant size (Bond and Van Wilgen, 1996; Watson *et al.*, 1997; Bond and Midgley, 2001; Matula *et al.*, 2012). Moreover, the replacement of stems by new sprouts without disturbances can benefit tree fitness through enhanced sexual reproductive output (Fujiki and Kikuzawa, 2006). Therefore, a wide range of extrinsic but also intrinsic factors may be driving sprouting in plants (Vila and Terradas, 1995a; Clarke *et al.*, 2010).

Despite the relevance of sprouting in population and community dynamics (Bond and Midgley, 2001, 2003; García and Zamora, 2003; Bellingham and Sparrow, 2009), little is known about the specific (either intrinsic or extrinsic) factors that trigger sprouting and the resulting multi-stemmed form in Iberian woody plant species. Most of the work has focused on common species and communities growing in Mediterranean environments affected by fires (Vila and Terradas, 1995a,b; Paula and Ojeda, 2006). On the contrary, few studies are focused on factors that might explain the inter-individual variability in sprouting at a microhabitat scale in endangered species or species growing at range margins in the Iberian Peninsula. An effort of this type would provide key information to understand the long-term survival of endangered or rare species

and would help to better design conservation programs (Bond and Midgley, 2001; García and Zamora, 2003).

The present study aims to discern what factors trigger different modes of sprouting and explain the inter-individual variability in the multi-stemmed architecture of the relict *Prunus lusitanica*. This understory tree is one of the subtropical species related to the Tertiary flora that persists in the Iberian Peninsula (García *et al.*, 2002). The progressive increase of seasonality and climate cooling that occurred during the second half of the Tertiary Era, and the Quaternary glaciations, are widely used to explain the current paucity and fragmented distribution of *P. lusitanica* in the Iberian Peninsula (Calleja, 2006). Extant populations are thought to be constrained to refuge habitats (mountainous and steep ravines) that buffer winter frosts and summer drought (Calleja and Sainz, 2009; García *et al.*, 2002). The rarity of this endangered species is also exacerbated by several human impacts that reduce population recruitment and suitable habitat (Calleja *et al.*, 2009).

To address which factors are relevant to sprouting in *P. lusitanica*, four different Iberian populations were selected at the Northeast edge of the global range of the species (Calleja, 2011). All studied populations live under similar climatic conditions (Table 1) that allowed us to explore to what extent non-climatic factors explain the variability in vegetative reproduction by means of basal sprouts and layering shoots. The factors and hypotheses considered are: (1) Tree size, which will determine the number of basal and layering shoots since larger sizes imply greater amounts of resources available for sprouting (Bazzaz *et al.*, 1987; Watson *et al.*, 1997; Bond and Midgley, 2001; Konsantinidis *et al.*, 2006). In addition, assuming a positive

**Table 1.** Sample sizes and climatic variables of the four studied populations of *Prunus lusitanica*

Pop	N° clumps	P sizes	Ha	Alt (m)	P (mm)	Pest (mm)	T <sup>a</sup>	T <sup>a</sup> max	T <sup>a</sup> min	Cont
LP	69	169	6.7	550	1.045	225	13	27.3	0.3	19.9
SPD	102	290	5.1	615	839	279	12.6	26.6	0.6	20.1
TM	100	277	5.9	790	881	280	11.7	24.9	0.6	18.2
CV	17	47	1.1	640	865	295	12.4	26.5	0.4	20.1

Pop: population. LP: Les planiques. CV: Can Verbolades. SPD: Sant Perè Desplà. TM: Turo de Morou. N° clumps: numbers of sampled clumps. P sizes: Population sizes (Calleja, 2011). Ha: area (measured in hectares) occupied by *Prunus lusitanica* populations. Alt: altitude, the average value of the altitudinal range of each population. P (mm): mean annual rainfall. Pest (mm): cumulative average rainfall from July to September. T<sup>a</sup>: mean annual temperature. T<sup>a</sup> max: mean maximum temperature of the warmest month. T<sup>a</sup> min: mean minimum temperature of the coldest month. Cont: Gorchynskis continentality index. This climatic data set is derived from a previous work (Calleja *et al.*, 2009).

relationship between size and age of individuals (Lieberman *et al.*, 1985; Boninsegna *et al.*, 1989; Matula *et al.*, 2012), a positive correlation between size and sprouting is expected since the occurrence of events that trigger sprouting might be cumulative over time. (2) Natural and anthropogenic disturbances (the fall, uprooting, breaking or logging of *P. lusitanica* trunks) could also trigger sprouting (Koop, 1987; Kammesheidt, 1999; Bond and Midgley, 2003). (3) Steep topography increases the likelihood of falling trees and branches that eventually root into the ground producing new shoots with vertical orientation (*i.e.* layering shoots). Thus, slope may promote multi-stemmed architecture in woody species (Nakamura and Inahara, 2007). (4) Canopy cover, such that closed canopies promote sprout bank development in several woody species pending the opening of gaps (Peterson and Jones, 1997; Tanentzap *et al.*, 2012). Therefore, we expect to find higher sprouting rates in shadier understories. (5) Soil moisture, such that sprouting, especially by means of layering shoots, is favored in wet environments (Koop, 1987; Del Tredici, 2001; Klimês *et al.*, 1997), therefore we expect to find the same relationship between soil moisture and sprouting in *P. lusitanica*.

## Material and methods

### Studied species

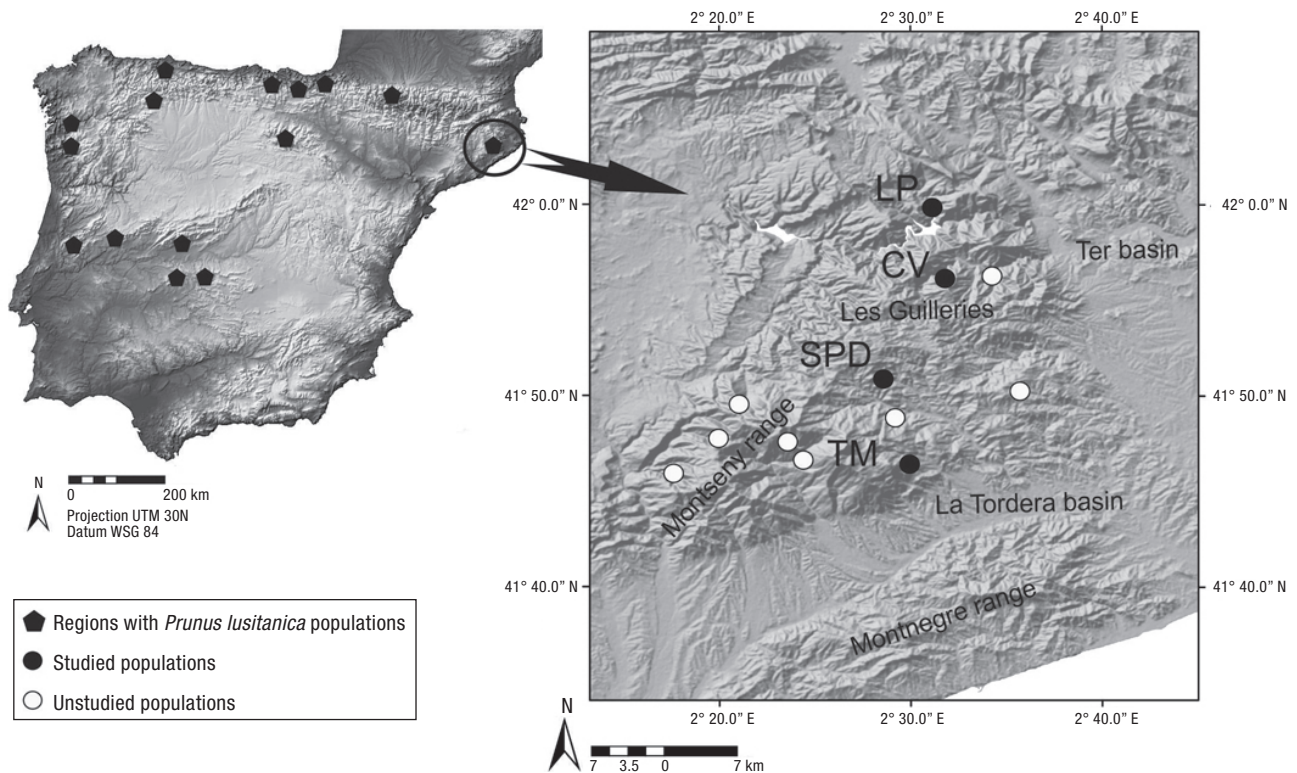
*P. lusitanica* is a broad-leaved evergreen tree, 3-18 m high. Its range includes the Iberian Peninsula, North Africa (Morocco's Rif Mountains) and the Macaronesian archipelagos of the Canaries, Azores and Madeira (Franco, 1964; Santiago, 2001). In the Iberian Peninsula, it usually grows on acid soils, living as an understory species within riparian communities along streams and beds near springs in closed mountain gullies, between 100-1,000 m.a.s.l. (Calleja and Sainz, 2009).

Although there is no fossil evidence, it is phylogenetically related to other subtropical cherry trees of Southeast Asia (Chin *et al.*, 2010). This fact plus its lauroid morphology and extant populations in the Macaronesian evergreen forest, indirectly support its consideration as subtropical relict species that inhabited the evergreen broad-leaved forests in the Mediterranean basin during the Tertiary (Pignatti, 1978; Barrón and Peyrot, 2006).

Today, *P. lusitanica* is recognized as a threatened species included in the Annex II of the Habitats Directive (Council Directive 92/43/EEC 1992) and has also been incorporated as "Vulnerable" in the national and regional catalogs of Spain (Calleja *et al.*, 2008; Aymerich *et al.*, 2010).

*P. lusitanica* exhibits two modes of reproduction: sexual reproduction through seeds and vegetative reproduction by means of basal and layering shoots (also called sprouts). Lateral sprouting roots or other vegetative reproduction modes have not been detected in the studied populations. *Basal shoots* arise from epicormic buds at the base of the main trunk and become secondary stems or trunks that share the same root system with their progenitor (Del Tredici, 2001). They promote self-regeneration and perpetuation of the individual (Bond and Midgley, 2001; Klimês and Klimês, 2007) but are not considered clonal propagation since they do not contribute to new independent offspring. *Layering shoots* arise from buds of lateral branches or trunks that come into contact with the ground and produce adventitious roots. These sprouts have their own root system but may remain indefinitely connected to their progenitors. Eventually, connections can fade out giving origin to new independent clonal individuals and thus, promoting clonal propagation. Both types of sprouts (basal and layering) are considered vegetative reproduction that promotes a multi-stemmed architecture (Calleja, 2006). The resulting *P. lusitanica* individuals are hereafter identified as clumps and can comprise four different stages: a) a single stem or trunk; b) a principal trunk with one to dozens of basal shoots sharing the same root system; c) a principal trunk with one to dozens of layering shoots; and d) a principal trunk with basal shoots and layering shoots. A population is defined as a group of clumps of *P. lusitanica* that live less than 1 km away from the closest clump of the same species.

The four selected populations of *P. lusitanica* are located at the Northeast of the Iberian Peninsula in the province of Girona (Catalonia), on the Massif of Montseny and Les Guilleries, within the basins of Tordera and Ter rivers (Fig. 1, Table 1). To ensure sufficient sample size for all the factors considered, the studied populations are the most numerous within the region (Calleja, 2011): Les Planiques, Sant Perè Desplà, Turó de Morou and Can Verbolades (Fig. 1, Table 1). The sampled clumps grow on the banks of small watercourses near springs. The complex topo-



**Figure 1.** Distribution of *Prunus lusitanica* in the Iberian Peninsula. The expanded graph shows the unstudied (white circles) and studied (black circles) populations in the NE range of the Iberian Peninsula: LP, Les planiques; CV, Can Verbolades; SPD, Sant Perè Desplà; TM, Turo de Morou.

graphy and steep slopes along with the sandy granites (with low cohesion), increase ground instability causing local landslides and falling trees. The average altitude is 650 m and the average annual temperature varies slightly around 12.5°C. The typical Mediterranean summer drought is poorly marked since the average summer rainfall is between 225 and 295 mm (Calleja, 2006) (Table 1).

The studied *P. lusitanica* clumps grow along with diverse temperate and Atlantic species (*Fraxinus excelsior* L., *Alnus glutinosa* (L.) Gaertn., *Corylus avellana* L., *Salix atrocinerea* Brot. and *Ilex aquifolium* L.). They are also occasionally mixed with some Mediterranean taxa, such as *Phillyrea latifolia* L. and *Arbutus unedo* L. The surrounding vegetation of the four studied populations consists of different evergreen and deciduous hardwood forests, which are distributed according to altitude, slope and soil moisture. Cork oak (*Quercus suber* L.) and holm oak (*Quercus ilex* L. subsp. *ilex*) forests grow in the lower and warmer areas. As altitude increases, these forests are enriched with pubescent oak (*Quercus pubescens* Willd.). In the higher and cooler areas, beech (*Fagus sylvatica* L.) becomes dominant.

## Data collection

The following data were collected from 288 clumps of *P. lusitanica*: (1) Number of basal shoots and layering shoots, as well as the total number of shoots including both types of sprouts and the principal trunk (*i.e.*, the largest trunk) of the clump. Sampled clumps were growing on different slopes or on different stream banks to avoid underground or layering connections among them. (2) DBH was calculated from the largest trunk perimeter, with an accuracy of 0.5 cm. DBH of the largest trunk is used as a surrogate of plant size. (3) The slope of the immediate surroundings was measured with a clinometer (Suunto PM-5) at the base of the main stem of each individual. (4) Soil moisture was categorized into two different classes by using different proxies within an area of one meter radius around each clump: i) wet: clumps growing on borders of streams and springs, or growing close to hygrophilous plants (according to Lara *et al.*, 2004, and Garilleti *et al.*, 2012): *Alnus glutinosa*, *Angelica sylvestris* L., *Carex pendula* Huds., *Carex remota* L., *Dryopteris affinis* (Lowe) Fraser-Jenk., *Fraxinus excelsior*, *Hypericum*

*androsaenum* L., *Luzula sylvatica* (Huds.) Gaudin, *Ranunculus repens* L.; ii) dry: elsewhere without the above characteristics. (5) Canopy cover; was categorized taking into account the size and length of gaps in a virtual circle (3-5 meters radius) centered above each clump. Gap sizes were estimated by visual inspection with the help of comparison charts of foliage cover (see supplementary material and William 1986). The time length of gaps was indirectly determined considering the deciduous or evergreen nature of the tree cover. We combined both types of data to establish two categories of canopy cover: i) closed canopy, where perennial tree cover over each studied clump exceeds 50% or when the tree canopy exceeds 75% and consists of both evergreen and deciduous trees (i.e. heavy shade from May to November); ii) open canopy: other coverage percentages and persistence regimes of tree cover above each clump. (6) The absence or presence of disturbances was recorded for each clump. Fallen main trunks or any of its secondary trunks and logging or breakages of any trunks within each clump were considered disturbances.

## Data analysis

The effects of the above factors (plant size, topography, soil moisture, canopy cover and disturbances), on the variability of sprouting ability were studied by constructing regression models in which the response variables were: (1) The total number of sprouts per clump, (2) the number of basal shoots per clump, and (3) the number of layering shoots per clump. The Generalized Linear Model (GLZ) with log link function was used assuming negative binomial distribution for response variables. The categorical explanatory variables were: soil moisture (wet or dry), canopy cover (closed or open canopy) and perturbation (presence or absence of disturbances). The slope and the size of the largest trunk of each clump were considered con-

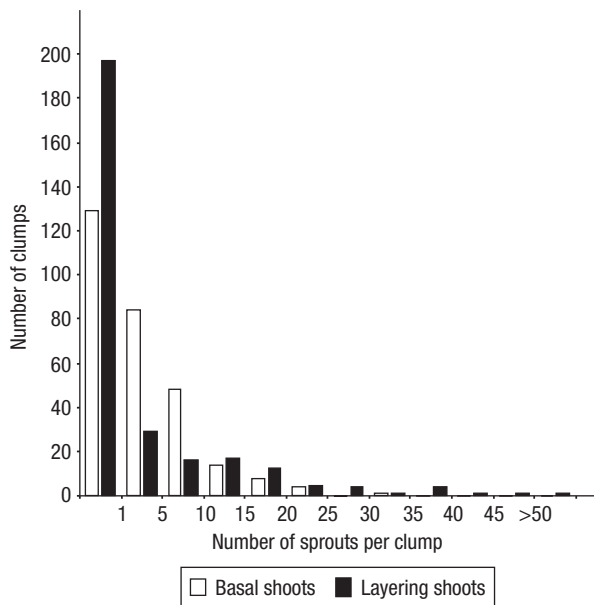
tinuous variables and included as covariates. In each model, the marginal means for the different levels of the categorical variables were estimated as the mean numbers of shoots (basal, layering and total shoots) calculated by setting the covariates at their respective means: 9 cm (for the largest trunk diameter) and 29° (for the slope). It was not possible to ensure the independence and orthogonality of all the variables considered. Likewise, we did not obtain a balanced number for all combinations of possible interactions between the different levels of the variables. Thus, the principal effects of the variables were statistically tested. In a complementary manner, the relationship between the number of sprouts per clump and the continuous variables (size and slope) were graphically and statistically explored by means of Pearson correlations. The potential effect of Size on the total number of sprouts at different levels of Perturbation was also graphically represented and analyzed by ANCOVA assuming homogeneity of slopes. Both size and the total number of sprouts were previously normalized by square-root and  $\log_{10}$  functions respectively. Then, both explanatory variables are represented without being normalized to facilitate the interpretation of the results. Additionally, we tested by ANOVA analysis the relationship between slope and the occurrence of fallen clumps of *P. lusitanica* (categorical variable). All the statistical analyses were carried out with SPSS 15.0.1 software (SPSS Inc., 2006).

## Results

The mean number of sprouts per clump is  $9.1 \pm 11.5$  (Table 2), although a large proportion of clumps have very few or no sprouts (Fig. 2). No remarkable differences have been found between the mean number of basal shoots ( $3.9 \pm 5.1$ ) and layering shoots ( $4.2 \pm 8.9$ ) per clump (Table 2). However, the maximum numbers of layering shoots per clump are greater or even double those of the basal shoots in most of the

**Table 2.** Mean, standard deviation, minimum and maximum numbers of basal shoots, layering shoots and total numbers of sprouts of the 288 sampled clumps of *Prunus lusitanica*

Population	Basal shoots		Layerin shoots		Total nº of sprouts	
Les Planiques	3.2 ± 4.6	0-20	4.0 ± 7.9	0-39	8.2 ± 10.5	1-49
Sant Perè Desplà	3.0 ± 3.9	0-19	4.9 ± 9.0	0-46	8.9 ± 10.4	1-49
Turó Morou	5.5 ± 6.3	0-33	4.3 ± 10.1	0-62	10.8 ± 13.6	1-85
Can Verbolades	2.6 ± 3.6	0-11	0.5 ± 1.0	0-4	4.1 ± 3.8	1-13
Total	3.9 ± 5.1	0-33	4.2 ± 8.9	0-62	9.1 ± 11.5	1-85



**Figure 2.** Frequency distribution of basal shoots (white bars) and layering shoots (gray bars), of the total number ( $n=288$ ) of clumps of *Prunus lusitanica* sampled in the four studied populations.

populations (Table 2). The highest numbers of basal shoots and layering shoots occur in the Turó de Morou population (Table 2).

The variables with a significant effect on the total number of sprouts in *P. lusitanica* are: the diameter of the main stem within each clump, disturbances, slope and canopy cover; however, soil moisture is not significant (Table 3). The number of shoots significantly increases as the diameter of the main stem is larger (Fig. 3a). In contrast, slope is poorly correlated with the production of sprouts (Fig. 3b). Canopy cover promotes significant differences in sprouting ability (Table 3), being higher under closed canopies, *i.e.*, in shaded environments, than in sunny ones (Table 4). Similarly, all of the disturbed individuals showed many

more sprouts than unaffected individuals, with values of  $12.2 \pm 1.8$  and  $5.7 \pm 0.4$ , respectively. In fact, the presence of disturbances invariably involves sprouting and is markedly more vigorous in larger trunks (Fig. 4), although there is not a significant interaction between perturbations and trunk size (Fig. 4).

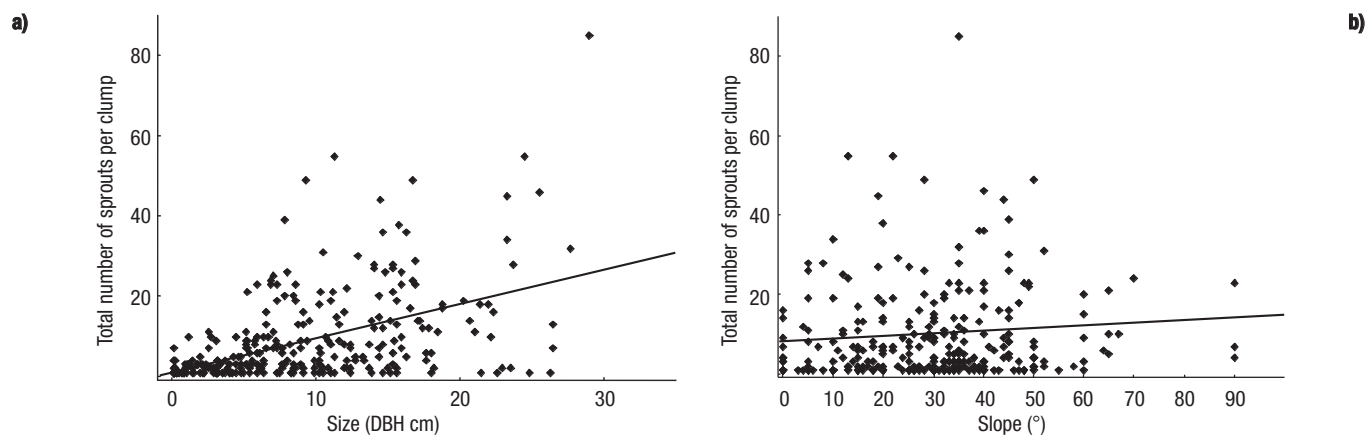
The diameter of the largest trunk within each clump is revealed as the main factor that positively influences the amount of both types of sprouts (Table 3). Similarly, disturbances trigger vegetative reproduction because their presence leads to an increase in sprouting (Fig. 4), especially by means of layering shoots (Table 4). Among the different types of disturbances, tree falling is the most common disturbance and causes a greater effect on the vegetative response, mainly in the production of layering shoots. Meanwhile, logging promotes a higher mean of basal shoots (Fig. 5). This type of sprout is also favored in shady environments (Table 4). On the contrary, slope and soil moisture only promote a significant change on layering shoot production (Tables 3 and 4). It is further noted that the proportion of fallen individuals is significantly greater on steeper

**Table 4.** Marginal means ( $\pm$  standard error) of sprouts for each level of explanatory variables, calculated by setting the covariates at their respective means: 9 cm in trunk diameter (*i.e.* size) and  $29^\circ$  of slope

Explanatory variables and levels	Total n° of sprouts	Basal shoots	Layering shoots
Perturbation			
No	$5.7 \pm 0.4$	$2.8 \pm 0.2$	$1.6 \pm 0.1$
Yes	$12.2 \pm 1.8$	$4.8 \pm 0.7$	$5.7 \pm 0.9$
Canopy cover			
Open	$7.2 \pm 0.9$	$2.9 \pm 0.4$	$2.7 \pm 0.4$
Closed	$9.8 \pm 0.9$	$4.6 \pm 0.4$	$3.4 \pm 0.3$
Soil moisture			
Dry	$7.6 \pm 0.9$	$3.8 \pm 0.5$	$2.3 \pm 0.3$
Wet	$9.2 \pm 0.9$	$3.6 \pm 0.4$	$4.0 \pm 0.4$

**Table 3.** Generalized Linear Model (GLZ) for the total number of sprouts, the number of basal shoots and layering shoots of the 288 sampled clumps of *Prunus lusitanica*

Explanatory variables	d.f.	Total n° of sprouts		Basal shoots		Layering shoots	
		Chi <sup>2</sup> Wald	<i>p</i>	Chi <sup>2</sup> Wald	<i>p</i>	Chi <sup>2</sup> Wald	<i>p</i>
Main trunk diameter	1	90.633	<0.001	63.768	<0.001	135.763	<0.001
Perturbation	1	21.427	<0.001	9.763	0.002	50.371	<0.001
Slope	1	9.156	0.002	2.088	0.148	23.326	<0.001
Canopy cover	1	4.999	0.025	8.578	0.003	2.105	0.147
Soil moisture	1	1.624	0.203	0.143	0.705	11.896	0.001



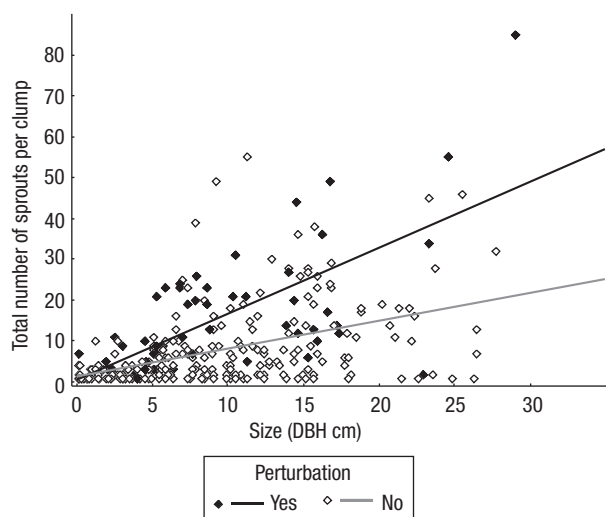
**Figure 3.** Pearson correlations between the total number of sprouts per clump of *Prunus lusitanica* and two quantitative variables: size (a) and slope (b). (a) Size ( $r=+0.53$ ,  $p<0.001$ ,  $n=288$ ) and (b) slope ( $r=+0.14$ ,  $p=0.015$ ,  $n=288$ ). Size (DBH) refers to the diameter of the largest trunk of each clump.

slopes (d.f. = 1,  $F = 4.127$ ,  $p < 0.05$ ), promoting the production of layering shoots (Fig. 6).

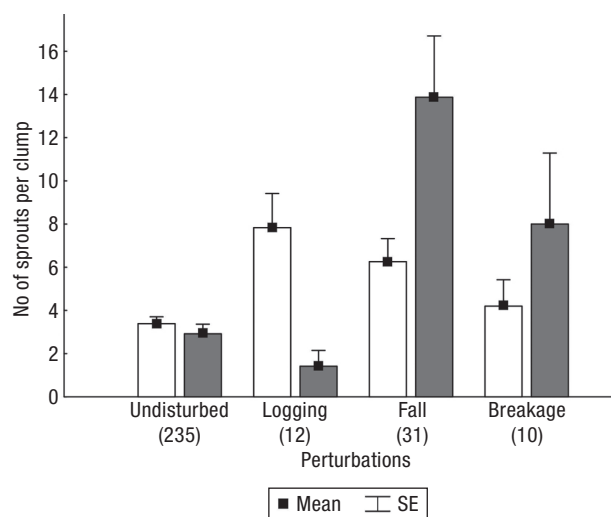
### Discussion

The multi-stemmed architecture of woody plants arises from sprouting that is often interpreted as an induced response to disturbances (Bond and Van Wilgen, 1996; Peterson and Jones, 1997; Bellingham and Sparrow, 2009). Sprouting, in the form of basal and layering

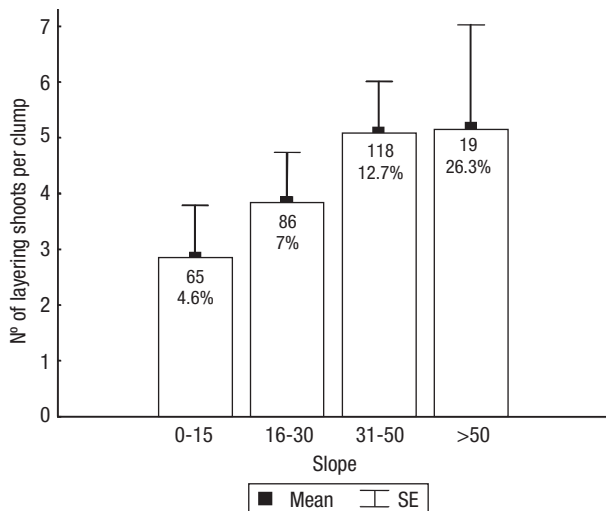
sprouts, in *P. lusitanica* is also promoted by several external (environmental and anthropogenic) factors but strongly influenced by the diameter (as a surrogate of plant size) of the largest trunk. The positive relationship between sprouting and plant size can be explained by two non-mutually exclusive factors. On the one hand, a larger diameter implies a potentially greater availability of investible resources in sprouting (Bazzaz *et al.*, 1987; Bond and Midgley, 2001; Konstantinidis *et al.*, 2006). Nonetheless, it does not mean that sprouting is a constitutive behavior linked to plant size in *P. lusitanica* since we found high numbers of single-



**Figure 4.** Effect of size (DBH) on vegetative reproduction of *Prunus lusitanica* with and without perturbations measured by ANCOVA analysis. Size (DBH) refers to the diameter of the largest trunk of each clump. Perturbation (d.f. = 1,  $F = 5.491$ ,  $p = 0.019$ ); Squareroot of Size (d.f. = 1,  $F = 58.478$ ,  $p < 0.001$ ); Perturbation x Size (d.f. = 1,  $F = 0.0002$ ,  $p = 0.989$ ); Intercept (d.f. = 1,  $F = 193.492$ ,  $p < 0.001$ ).



**Figure 5.** Effect of different types of perturbations on sprouting. Mean (black dots) and standard error (SE, whiskers) are shown for basal shoots (white bars) and layering shoots (gray bars). Sample sizes within each perturbation class are shown between brackets.



**Figure 6.** Number of layering shoots per clump of *Prunus lusitanica* with fallen trunks growing on different slope ranges. Total number of sampled clumps and respective percentages of clumps with fallen trunks are shown within each slope class box.

stemmed clumps irrespective of the diameter of their trunks. On the other hand, plant size is usually positively correlated to the age of the aboveground parts of plants (Lieberman *et al.*, 1985; Boninsegna *et al.*, 1989; Matula *et al.*, 2012). A larger size could mean a longer life history, and therefore could increase the likelihood of sprout generation triggered by different external factors. Natural or anthropogenic disturbances can add up over time and, indeed, it appears that the studied cases (tree falling, broken trunks and logging) promote sprouting. Therefore, larger trunks would accumulate greater sprout banks throughout time even if the sprouting ability decreases with age (Sands and Abrams, 2009; but see Matula *et al.*, 2012). Thus, plant size does not trigger sprouting but it primarily drives the size of the sprout bank (in terms of number of sprouts) in *P. lusitanica*.

All of the external factors considered promote sprouting in *P. lusitanica* but differ on their effect on the numbers of the two types of studied sprouts. Basal shoots are mainly triggered by some natural (broken and fallen trunks) or anthropogenic (logging) perturbations as occurs in several woody species (Kammesheidt, 1999; Nagaike *et al.*, 1999). The sprouting ability by basal shoots reveals some potential of *P. lusitanica* to persist by self-regeneration despite some natural perturbations and human threats (Kammesheidt, 1999; Nagaike *et al.*, 1999; Bond and Midgley, 2001; García and Zamora, 2003; Bellingham and Sparrow, 2009; Tanentzap *et al.*, 2012). Canopy cover also affects the

production of basal shoots. *P. lusitanica* clumps growing in shadier understories possess higher numbers of basal sprouts like many other subtropical evergreen broad-leaved species (Fernández-Palacios and Arévalo, 1998; Kobe, 1999). The possession of a sprout bank in poor light environments improves the chance of rapid vegetative reproduction when a gap occurs in the forest canopy (Hara, 1987; Peterson and Jones, 1997). In the long term, the sprouting ability of *P. lusitanica* by basal shoots might prolong the lifespan of clumps under stressful environments and after some perturbations (Del Tredici, 2001; Bellingham and Sparrow, 2009; Tanentzap *et al.*, 2012) and may contribute to conserve the colonized habitat (Bond and Midgley, 2001; Kawecki, 2008).

The development of layering shoots of *P. lusitanica* clumps is a consequence to the combination of rugged topography and soil moisture. The steep slopes along with local landslides promote frequent uprooting and falling of principal and secondary trunks that produce layering shoots from the branches that contact the ground as seen in unstable habitats (Koop, 1987; Nakamura and Inahara, 2007). Moreover, the number of these layering shoots significantly depends on soil moisture (see also Del Tredici, 2001) inherent to the streams and springs around which *P. lusitanica* usually grows (Calleja and Sainz, 2009). Thus, compared to basal shoots, layering shoots depend on the simultaneous action of two different external factors (topography and soil moisture) to succeed. Over time, connections between layering shoots and their progenitors may die away. Therefore, these sprouts not only allow for self-regeneration of clumps, as basal shoots do (Bond and Midgley, 2001; Tanentzap *et al.*, 2012), but also promote clonal propagation and local expansion since they grow at measurable distances from the progenitor trunk.

In summary, the multi-stemmed architecture of *P. lusitanica* at the Northeastern limit of its worldwide distribution is driven by local environmental factors (canopy cover, slope and soil moisture) and disturbances, both natural and anthropogenic. Each external factor shows different levels of influence on the variability and type of vegetative reproduction (basal and layering shoots) yet the intensity of the response is principally influenced by the size of the largest trunk of each clump. Layering shoots promote local expansion and both types of sprouts favor the resilience of the species against some stressful conditions thereby contributing to the persistence of populations of *P. lusitanica* in the Iberian Peninsula. However, to truly



understand this contribution to the long term survival of the species, it is necessary to undertake a detailed and prolonged population dynamics study that involves the growth and survival rates of sprouts (Cornelissen *et al.*, 2003). It would also be worthwhile to carry out a larger scale comparative study of sprouting within different geographic and climatic scenarios (Macaronesia and Iberian Peninsula). This would help to clarify the relevance of sprouting to the survival of *P. lusitanica* in adverse climates and to what extent sprouting contributes to the conservation of the ecological niche and geographic distribution over evolutionary time.

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