New Forests
https://doi.org/10.1007/s11056-020-09818-x

The use of biological liquid fertilizers against oak decline associated with *Phytophthora* spp.

Aida López-Sánchez[1] · Ramón Perea[1]

Received: 27 July 2020 / Accepted: 5 November 2020

Abstract

Worldwide, multiple ecosystems are currently affected by global change because of important anthropogenic disturbances which have quickly increased levels of abiotic (e.g. climate) and biotic stress (e.g. diseases, pests, herbivory). In particular, oak-dominated systems have experienced a general decline as a consequence of habitat destruction, mismanagement and extreme climate events along with the aggressive root pathogen *Phytophthora* spp. Here, we investigated the effect of soil improvement through soil biological fertilizers on oak defense against tree decline symptoms associated with *Phytophthora* for two ontogenetic stages of oak trees: adults and recruits. We examined oak survival and crown defoliation status on 60 pairs of adult trees (a treated tree with biological fertilizers vs. non-treated tree) as well as the survival, growth and herbivory on 30 quartets of treated and non-treated recruits, with and without protection against large herbivores. We used two different liquid fertilizers: OptiPlus (a biological fertilizer with organic N, P, S, Ca, Mg, trace elements (Fe, Mn, B, Zn) and humic fulvic acids) and OptiFer (a biological trace element fertilizer with only Fe, Mn and Mg). Treated oak trees with fertilizers showed lower oak defoliation (0.33-fold difference) than non-treated trees. In addition, trees treated with OptiPlus liquid showed a significant reduction of crown defoliation compared to those trees that received OptiFer treatment (0.68-fold difference). Interestingly, this crown amelioration was more effective in steep slopes, revealing that OptiPlus may buffer the stress generated by shallower and poorer soils. Overall, fertilized plants (particularly OptiPlus) were more attractive to herbivores, probably as a result of the greater content in deficient minerals, increasing their palatability. This study has slightly visualized the importance of soil improvement against oak decline through biological fertilizers and the need to further explore different times and techniques of application. Managers should be cautious with the use of fertilizers as they may increase the browsing impact of livestock and wild herbivores on tree regeneration. It is therefore imperative to improve the resilience of oak trees against oak decline while minimizing the herbivory impact on oak recruits.

Keywords Biological fertilizers · Tree crown defoliation · Oak regeneration · Herbivory · *Phytophthora cinnamomi* · Soil quality

Electronic supplementary material The online version of this article (https://doi.org/10.1007/s11056-020-09818-x) contains supplementary material, which is available to authorized users.

Extended author information available on the last page of the article

Introduction

Worldwide, multiple ecosystems are currently affected by global change, as a consequence of important anthropogenic disturbances (e.g. land-use changes, socio-economic and climate disruption, species introductions; Ellison et al. 2005; Aukema et al. 2010; Metz et al. 2017). Since mid-20th century, these disturbances have quickly modified abiotic (e.g. climate) and biotic factors (e.g. diseases, pests, herbivory) increasing their levels; and consequently, generating new sources of stress for plants (Blondel and Aronson 1999; Underwood et al. 2009; Metz et al. 2017). For instance, water deficit and temperature increase—new sources of abiotic stress strengthened by anthropogenic climate change (Lutz et al. 2010; IPCC 2014), have favored the arrival and spread of emerging diseases and pests (abiotic-biotic interaction of stress sources). These interactions between stress agents have increased the decline and even the mortality of many species in all their life stages (Brasier et al. 1993a; Haavik et al. 2015; Serrano et al. 2017). In addition, unprecedented levels of herbivory have been documented in many systems, mostly due to the increase of wild ungulate populations (Gordon et al. 2004; San Miguel et al. 2010) and the unsuitable rangeland management practices (Pulido et al. 2001; Plieninger et al. 2003; López-Sánchez et al. 2017). Consequently, the structure and functioning of many ecosystems might be compromised if the different biological components do not keep pace with the disturbance cascade (White and Jentsch 2001; Johnstone et al. 2016).

Oaks (*Quercus* spp.) are a dominant tree component across the Northern Hemisphere natural systems, which provide important ecosystem services (Caparrós et al. 2013). However, nowadays, oak-dominated systems are severely affected by increasing sources of stress (e.g. drought, diseases, herbivory). For instance, 'la seca', the oak decline due to abiotic-biotic interaction of stress sources (Duque-Lazo et al. 2018), is threatening the oak survival in many oak woodlands (Brasier 1993; Rizzo and Garbelotto 2003; Campos et al. 2013; Jung et al. 2018). Several pathogens including the oomycete *Phytophthora cinnamomi* (soil-borne root pathogen causing the ink disease), are important drivers triggering oak decline (de Sampaio e Paiva Camilo-Alves et al. 2013; Hansen 2015; Ruiz-Gómez et al. 2019) along with habitat destruction, mismanagement and extreme climate events (Branco and Ramos 2012; Duque-Lazo et al. 2018; Gentilesca et al. 2017). The result of oak decline is a strong oak growth reduction and a progressive degradation of canopy foliation and root systems (Haavik et al. 2015). Furthermore, oaks are not natural host-plant robust to *P. cinnamomi* (Serrano et al. 2012a), and most of the oaks affected by *Phytophthora* spp. usually die after their infection (Brasier et al. 1993b; de Sampaio e Paiva Camilo-Alves et al. 2013; Haavik et al. 2015). On the other hand, the increased herbivory pressure on oaks, especially over young plants, is considered an important driver of the current regeneration failure in most oak woodlands worldwide (Pulido and Díaz 2005; Tyler et al. 2006). Previous research has shown that high densities of ungulates (wild or domestic) trigger severe problems on plant regeneration (Côté et al. 2014; López-Sánchez et al. 2016; Charro et al. 2018). In addition, drier conditions exacerbate the herbivory intensity on oaks, especially on oak regeneration (Zamora et al. 2001; López-Sánchez et al. 2019). The stressing effect of herbivory could produce instability on the performance and development of plants during their growth (Møller and Shykoff 1999; Nolte 2003). In some cases, repeated herbivory on plants may even cause their death (Nolte and Dykzeul 2000; López-Sánchez et al. 2014; Perea and Gil 2014).

Applied research is, therefore, critical in order to find solutions that minimize the effects of these new sources of stress. Some researchers have found temporal solutions (e.g. soil fertilizers, cultural actions) to refrain oak decline affection due to *Phytophthora* spp. (Serrano et al. 2012b, 2017). For instance, elevating calcium content in soil has been proved to supress chlamydospore viability of pathogenic oomycetes (Broadbent and Baker 1974; Duvenhage et al. 1992; Heyman et al. 2007; Serrano et al. 2012b, 2017). Cultural actions such as keeping a correct soil drainage, maintaining an appropriate livestock stocking rate, or limiting soil tillage and movements from vehicles or livestock have demostrated to reduce the *Phytophthora* spread (Fernández et al. 2008; Serrano et al. 2017). Fungicides for *Phytophthora*, in contrast, were found not effective in *Q. ilex* population (Navarro et al. 2009); however, biofumigation (e.g. on *Brassica* species) has shown to inhibit effectively mycelial growth of *P. cinnamomi* (Dunne et al. 2003; Ríos et al. 2016; Serrano et al. 2017).

In addition, fertilizers, in general, contains important deficient minerals for both plants and animals (Gambín et al. 2017). Thus, fertilizations increase the palatability of plants for the main herbivores as a result of the increased nutrient content of the fertilized plants (Oh et al. 1970; Burney and Jacobs 2013). In general, ungulates selectively browse plants with higher contents of protein and energy (i.e., carbohydrates) and avoid plant with higher contents of toxins (Provenza et al. 2003). Thus, highly palatable plants are intensively browsed, sometimes threatening their conservation (Miranda et al. 2011; Fernández-Olalla et al. 2016). Consequently, it is important to find solutions to improve the resilience of oaks against oak decline that minimize the herbivory impact on oaks, in particular on oak regeneration as it is a preferred browse in many systems (Perea et al. 2014, 2020).

In this study, we investigate the effect of soil improvement through soil biological fertilizers on oak defense against decline symptoms for two ontogenetic stages of oak trees: adults and oak recruits. We specifically hypothesized that: H1) oak tree survival and health (crown defoliation status) will increase after the application of the biological liquid fertilizers; H2) survival and growth of oak recruits will also increase after the fertilizer application; and H3) treated oak recruits will be more heavily browsed than control recruits as a result of the increased nutrient content of the fertilized plants. We tested these hypotheses in Mediterranean oak woodlands of Spain, which are recognized for their ecological, socioeconomic and cultural importance (Plieninger and Wilbrand 2001; Dagit et al. 2015; López-Sánchez 2015). Hence, the final aim of this study was to search for applied measures that invigorate trees and recruits, allowing them to better resist oak decline virulence and other threatening drivers; and thus enhance the conservation of these valuable ecosystems.

Materials and methods

Study area

The study was conducted over a year (November 2018–October 2019) in a traditionally managed scattered oak system. The "Dehesa de San Francisco" (515 ha) is located in Huelva province (Spain) in the Southwest of the Iberian Peninsula (37°52′ N, 6°14′ W; 352–528 m a.s.l; Fig. 1) and has a strong slope variability (10–80%; being 20–35% the most frequent slopes). The climate is Mediterranean pluviseasonal oceanic, with hot and dry summers. Mean annual temperature is 17.2 °C and mean annual rainfall is 677.1 mm (n = 21 years; weather station IQ102 "Santa Olalla de Cala" 37° 54′ N, 6° 13′ W), concentrated in winter months and with strong interannual variability (5.43–128 mm/month). Annual rainfall of the study year was 326.5 mm. Parent material of soil formation at the research area is schist. Due to the strongly dissected topography, some soil material has

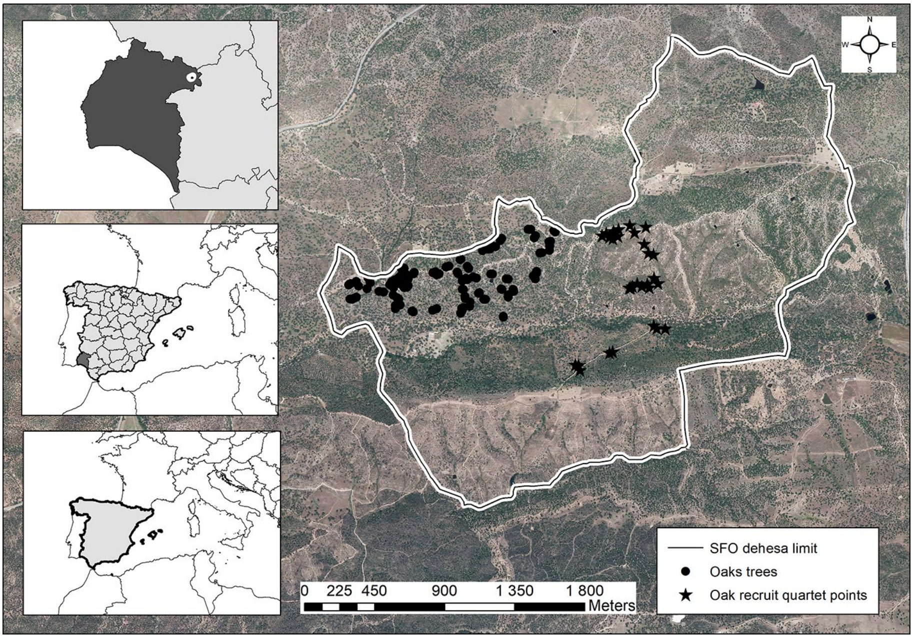

Fig. 1 Map of study area location, and the oak adult and oak recruit quartet location

moved down the slopes and is forming colluvic material (IUSS 2014) at the footslopes. Predominantly, continuous rock (schist) starts ≤ 25 cm from the soil surface. Hence, these soils belong to the Reference Soil Group (RSG) of Leptosols (IUSS 2014).

The tree layer is co-dominated by holm oak (*Quercus ilex* subsp. *ballota* (Desf.) Samp.) and cork oak (*Quercus suber* L.), locally interspersed with gall oaks (*Q. faginea* subsp. *broteroi* Cout. A.Camus). Mean tree density is around 82 trees ha^{-1}. The shrub layer is low and dominated by evergreen xerophytes (e.g. *Cistus salviifolius* L., *Cistus ladanifer* L., *Lavandula stoechas* Lam., and *Genista hirsuta* Vahl). The herbaceous layer mainly comprises therophytic oligotrophic communities (*Tuberarietalia guttatae)* and sub-nitrophilous Mediterranean annual communities *(Thero-Brometalia)*; see Rivas-Martínez et al. (2001). Grasslands belonging to the *Poetea bulbosae* community are also common; they are promoted by intense and continuous livestock grazing and dominated by small perennial grasses and nutritious forbs (Rivas-Martínez et al. 2001).

A traditional rotational grazing system with typical dehesa livestock species (cattle, sheep, goats, and Iberian pigs) has been re-established from 2004 onwards, and the farm has been subjected to organic farming since then. The cattle breeds "Retinta", "Berrenda en rojo", "Berrenda en negro", and "Limousin" are kept year-round on the farm with stocking rates of 0.042 LU ha^{-1}. Sheep (mainly the breed "Merina") are kept with stocking rates of 0.104 LU ha^{-1}, while goats have stocking rates of 0.004 LU ha^{-1}. The pig breed is "Ibérica" with stocking rates of 0.090 LU ha^{-1}. In addition, wildlife such as red deer (*Cervus elaphus* L.), fallow deer (*Dama dama* L.) and wild boar (*Sus scrofa* L.) are present on the farm with overall densities of ca. 0.081 LU ha^{-1}.

Our study area is located in Andalusia region, in which, since mid-20th century, oaks have exhibited stress that usually ends up dying and it is usually associated with *P. cinnamomi* (Sánchez et al. 2002; Duque-Lazo et al. 2018). The study area is severely affected by ‘Seca’ (25% of Dehesa, owners, pers. comm; Fig. S1). Several pathogens including *Phytophthora cinnamomi* and *P. andina* are present in the soil which are severely affecting oak trees in the study area (Sapp et al. 2019).

Description of the products

We used two different liquid products: OptiPlus and OptiFer (Optima-Werker Biologische Produkte für Haus und Garten, Münchenstein, Switzerland). OptiFer is a biological trace element fertilizer that contains 6% Fe, 1.5% Mn, 1.5% Mg and the remainder is water. The trace elements are extracted from the bark of trees with *Tsuga canadensis* as the main source. Their effect is to produce the symbiosis between plants and soil organisms through roots. Soil organisms transfer trace elements into the hair roots of the trees. OptiPlus is a biological humic fertilizer that contains 0.8% N organic, 0.3% P, 0.2% P organic, 0.5% S, 1% Ca, 0.18% Mg, 0.3% trace elements (Fe, Mn, B, Zn), 4.5% organic substance, 20% humic fulvic acids, and the remainder is water. Humic substance is an effective nutritional fertilizer without NPK, which is immediately receptive to the plants. Trace elements of both fertilizers travel from roots until leaves boosting chlorophyll formation. In addition, this trace element enrichment help to expand cell walls of the leaves increasing the sunscreen. The effects of “thicker” leaves is the resistance to pests and aggressiveness of destructive fungi of all kinds (Jones and Dangl 2006; Yang et al. 2018).

Data collection

Oak trees

In November 2018, we selected 120 adult holm oaks (*Q. ilex*) in a site (107 ha) severely affected by oak decline (Fig. 1; Fig. S1) where *Phytophthora cinnamomi* was recorded (Sapp et al. 2019). Within the site, we selected trees in pairs (approximately 5–20 m between trees). Each pair comprised two holm oak trees with very similar size (similar diameter at breast height, dbh at 130 cm high), ecological conditions (distributed on the same hillside following a slope gradient), and crown damage. Only trees clearly affected (> 10% crown defoliation) were selected for the experiment to ensure that damage is not only due to possible insect defoliation, which typically affect < 10% of the crown biomass. In addition, we avoided pairs with more than 90% crown defoliation since they are almost dead.

One of the trees within each pair received a liquid treatment and the other was the control. We applied the product in a season without drought (November 2018). Thus, we assure that improvement of tree condition is due to product and not due to the “needed” water contribution. A total of 60 trees received treatment by watering gently 100 L (30% product + 70% water) of mix on the base of the trunk from the uphill side of the tree to avoid liquid losses as less as possible. Thirty trees received OptiPlus liquid, 30 trees received OptiFer liquid and 60 acted as control.

Over the study year, in each tree, we measured the following variables twice (before and after liquid product application): dbh (diameter at breast height), total crown diameters, life crown diameters and crown defoliation. Total crown diameters are those that include all branches, both foliated and completely defoliated. For each tree, we measured the longest total crown diameter (d_1) and its perpendicular (d_2), and calculated the total crown area. We also measured life crown diameters, which are those that include only foliated branches, and calculated the life crown area. We obtained the variable crown area loss as the percentage of life crown area in relation to the total crown area. Crown defoliation was standardized by estimating the percentage of crown defoliation in comparison with a fully foliated (healthy) reference tree of the same species (Müller and Stierlin 1990). Physiographical variables were also collected for each tree (e.g. slope, aspect) using satellite images.

We, then, calculated the difference of crown defoliation and crown area loss between the two visits (November 2018 and October 2019), obtaining crown defoliation rate and the crown area loss rate (response variables).

Oak recruits

In November 2018, we selected 120 oak recruits (dbh < 5 cm) grouped in quartets (total: 30 quartets) of two different oak species—holm and cork oak (*Q. ilex* and *Q. suber*, respectively)—in a site (30 ha) where *Phytophthora cinnamomi* is widespread (Sapp et al. 2019; Fig. 1). Each quartet (approximately 25–100 m^2) contained four oak recruits of the same species and with very similar size (basal diameter and crown), ecological conditions (distributed on the same hillside and fairly close to each other, 4–10 m for most recruits), and crown damage. However, most of the oak recruits did not have any apparent damage; thus, we selected all of them with similar defoliation (mostly light damage; 10–30% defoliation due to the lack of oak recruits with heavier damage). When light-damaged recruits were rare, we used healthy recruits to analyze the effect of the product in preventing future possible damage since *Phytophthora* spp. is present in the study area (Sapp et al. 2019). A total of 60 recruits received OptiPlus treatment by watering gently 50 L (30% product + 70% water) of mix on the base of the recruit from the uphill side of the tree to avoid liquid losses as less as possible. The other 60 recruits acted as control. In addition, 60 recruits (one treated and one control per quadrat) were protected against ungulates with wire cages (2 m high) and the rest of 60 recruits were not protected. Control recruits were always located uphill of the treated recruits to avoid product invasion.

Over the study year, for each oak recruit, we measured the following variables twice (before and after liquid product application): basal diameter, plant height and plant crown diameters. For crown diameters, we measured the longest total crown diameter of each recruit (d_1) and its perpendicular (d_2). We also recorded ungulate herbivory damage. For that, we selected ten top twigs of the recruit and marked those browsed by herbivores obtaining a damage percentage which might be categorized following Perea et al. (2015). We, then, calculated the difference of plant size (recruit height and crown area) and herbivory between two visits (November 2018 and October 2019) obtaining plant survival, plant height rate (hereafter growth rate), recruit crown area rate and herbivory rate (response variables).

Data analysis

We developed several maximal Generalized Linear Mixed Models -GLMMs- (Venables and Ripley 2002) to analyze the data. The maximal models (containing all predictors) are summarized in Table 1. Box–Cox transformations (Box and Cox 1964) were applied when needed in order to calculate the lambda transformation that maximizes the likelihood. Thus, some of the response variables were fitted to Gamma and Binomial error distribution with their corresponding power lambda link function (Table 1). When monotonic transformations were not necessary, the response variables were fitted to Gaussian error

Table 1 Summary of maximal Generalized Linear Mixed Models performed for data analysis in this study

<table>
<tr><th>Layer</th><th>Model</th><th>Response variable</th><th>Fixed effect[a]</th><th>Random effect</th><th>Error distribution (power lambda link function)[b]</th><th>Sample size (n)</th></tr>
<tr><td>Oak trees</td><td>I</td><td>Crown defoliation rate</td><td>T × S + T × A</td><td>1|Pair</td><td>Gamma (0)</td><td>120</td></tr>
<tr><td></td><td>II</td><td>Crown defoliation rate</td><td>P × S + P × A</td><td>1|Pair</td><td>Gamma (0)</td><td>120</td></tr>
<tr><td></td><td>III</td><td>Crown area loss rate</td><td>T × S + T × A</td><td>1|Pair</td><td>Gaussian (1)</td><td>120</td></tr>
<tr><td></td><td>IV</td><td>Crown area loss rate</td><td>P × S + P × A</td><td>1|Pair</td><td>Gaussian (1)</td><td>120</td></tr>
<tr><td>Oak recruits</td><td>V</td><td>Oak survival</td><td>T × Sp + T × Pr</td><td>1|Quartet</td><td>Binomial ()</td><td>120</td></tr>
<tr><td></td><td>VI</td><td>Herbivory rate</td><td>T × Sp + T × Pr</td><td>1|Quartet</td><td>Gamma (−0.263)</td><td>120</td></tr>
<tr><td></td><td>VII</td><td>Plant growth rate</td><td>T × Sp</td><td>1|Quartet</td><td>Gamma (0.250)</td><td>60</td></tr>
<tr><td></td><td>VIII</td><td>Recruit crown area rate</td><td>T × Sp</td><td>1|Quartet</td><td>Gamma (−0.101)</td><td>60</td></tr>
</table>

[a]Oak trees = T: Treatment (control vs. treated); P: Product used in the treatment (control, OptiPlus vs. OptiFer); S: Slope; A: Aspect

Oak recruits = T: Treatment (control vs. OptiPlus); Sp: Species (cork oak vs. holm oak); Pr: Protection (fenced vs. non-fenced)

[b]Power lambda link function [$g(\mu) = \mu^{\lambda}$] is the lambda (λ, numeric value inside the brackets) used for the monotonic transformations

distribution with identity function. For all models, the analyses included each "plot" (tree pair or recruit quartet) as the random effects structure (Table 1).

We used the model averaging approach (Burnham and Anderson 2002). We first fitted the maximal model, containing all predictors. Then, we ranked through AIC weights all possible models derived from the maximal model by using the "dredge" function within the "MuMIn" package of R and selected those with the highest AIC weight (hereafter top models) which had $\Delta AIC < 2$ (Burnham and Anderson 2002). Finally, we obtained the model-averaged coefficients of top models as well as the relative importance of each predictor (from 0 to 1) by using the "model.avg" function of "MuMIn" (Burnham and Anderson 2002).

Data processing and statistics were performed using R 3.6.0 (R Development Core Team 2019) with the modules "lme4" (Bates et al. 2015), "car" (Fox and Weisberg 2011), "MuMIn" (Barton 2015).

Results

Oak trees

Only one adult oak tree died over the study period. We found differences in crown defoliation rate (models I and II) depending on treatment (control vs. treated) and type of product (Table 2). Control trees (without treatment) had higher crown defoliation rate than those treated trees with liquid product (3.01-fold difference; Fig. 2i). In addition, trees treated with OptiPlus liquid showed a significant reduction of crown defoliation rate compared to those trees that received OptiFer liquid product (0.68-fold difference; Fig. 2ii).

On the other hand, we only detected significant differences in crown area loss rate (models III and IV) depending on treatment and type of product when interacting with slope (Table 2). In the case of low slope (0°–10°), we did not find any significant differences in crown area loss rate among treated and control trees (Fig. 3). However, as the slope increased, the crown area loss rate also increased for controls whereas, for treated trees, the soil liquid treatment applied reduced the stress generated by the slope increase (Fig. 3). We did not find any significant differences for trees treated with OptiFer liquid product interacting with slope because most of trees that received OptiFer liquid treatment had similar slope (0 to 10 degrees; Fig. 4).

Oak recruits

The survival of young oaks was very high (97%). Hence, we did not find significant differences in survival depending on product type ($z = 0.004$, $P = 0.997$), species ($z = 0.004$, $P = 0.997$), or herbivore protection ($z = 1.060$, $P = 0.284$).

We found that holm oaks showed in general less herbivory than cork oaks (0.81-fold difference; Table 3, Fig. 5i); however, herbivory was higher on holm oaks when OptiPlus was applied on recruits (7.94-fold difference compare to controls) than for cork oaks treated with OptiPlus (1.23-fold difference compare to controls; Fig. 5i). We did not find significant differences in plant herbivory rate (model VI) depending on product itself (Table 3). In addition, for protected oaks, herbivory levels increased for OptiPlus treated-oaks compared to controls (7.94 fold difference), but they still had lower herbivory damage than non-protected oaks (0.27-fold difference; Fig. 5ii).

Table 2 Summary of the top linear mixed models (ΔAIC < 2) to analyze the crown defoliation rate (I and II models) and crown area loss rate (III and IV models) depending on treatment and product (covariates: slope and aspect are also included)

Model	Fixed effects	*Importance*	Levels	*Coeff.*	*SE*	*z-value*	*P*
I	Intercept			0.694	0.270	2.543	0.011
	Treatment (T)	1.00	Liquid	– 1.049	0.191	5.443	<0.001
	Slope (S)	0.41	Slope	0.116	0.145	0.791	0.429
	Aspect (A)	0.55	Aspect	0.226	0.175	1.280	0.201
	T × S	0.18	$T_{Liquid} \times S$	– 0.159	0.204	0.772	0.440
II	Intercept			0.729	0.268	2.687	0.007
	Product (P)	1.00	OptiFer	– 0.656	0.232	2.800	0.005
			OptiPlus	– 1.647	0.289	5.640	<0.001
	Slope (S)	0.43		0.173	0.159	1.077	0.282
	Aspect (A)	0.45		0.106	0.140	0.752	0.452
III	Intercept			35.276	4.707	7.397	<0.001
	Treatment (T)	0.87	Liquid	7.015	6.983	0.994	0.320
	Slope (S)	0.66	Slope	0.631	0.333	1.855	0.064
	T × S	0.51	$T_{Liquid} \times S$	– 1.107	0.509	2.129	0.033
IV	Intercept			34.132	4.010	8.512	<0.001
	Product (P)	1.00	OptiFer	8.388	6.564	1.278	0.207
			OptiPlus	13.742	7.912	1.737	0.088
	Slope (S)	0.82	Slope	0.568	0.322	1.763	0.084
	P × S	0.75	$P_{OptiFer} \times S$	– 0.449	0.612	– 0.734	0.466
			$P_{OptiPlus} \times S$	– 1.818	0.713	– 2.549	0.014

Importance: Importance of predictor variable in the model averaging. Results from Treatment and Product type are against control trees

We did not find any significant differences in plant size (growth rate -model VII- and crown area rate -model VIII-) depending on product or species (Table 3). However, holm oak recruits showed marginally lower growth rates compared to cork oak recruits ($P = 0.071$; Table 3).

Discussion

Our results highlight that both increasing sources of stress (*Phytophthora* and herbivory) which are affecting many oak woodlands worldwide, are widespread in the study area but apparently affect ontogenic stages differently, with adults being mostly associated with *Phytophthora* symptoms and the recruitment mainly affected by high levels of ungulate herbivory. Here, we proved the efficacy of some biological fertilizers in reducing the oak decline symptoms under certain conditions (hypotheses H1 and H2) although it may increase the damage by other sources of stress such as herbivory on young plants (H3).

Previous research has shown that soil fertilizers might be a possible solution to eradicate or significantly reduce the oak decline effects (Serrano et al. 2012b, 2017); however, further research is really needed in this regard. Our results have shown that biological fertilizers may improve soil conditions and reduce oak decline (H1). Furthermore, over the

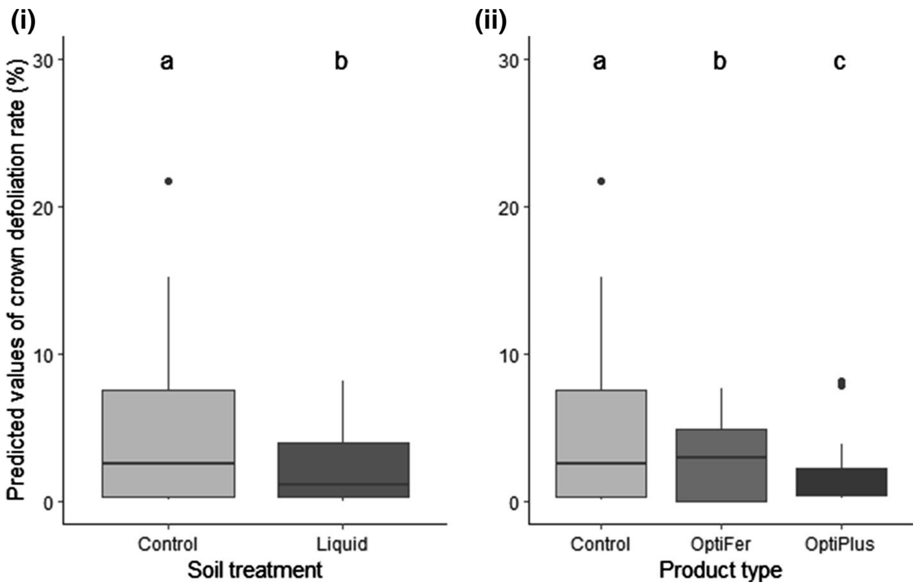

Fig. 2 Predicted values of crown defoliation rate (%) depending on (i) treatment and (ii) liquid product (N = 120 trees). Control = trees within each pair that received no liquid treatment, Liquid = trees within each pair that received liquid treatment. OptiFer = trees that received OptiFer liquid treatment, OptiPlus = trees that received OptiPlus liquid treatment. Different letter above the boxes indicate significant differences ($P < 0.05$)

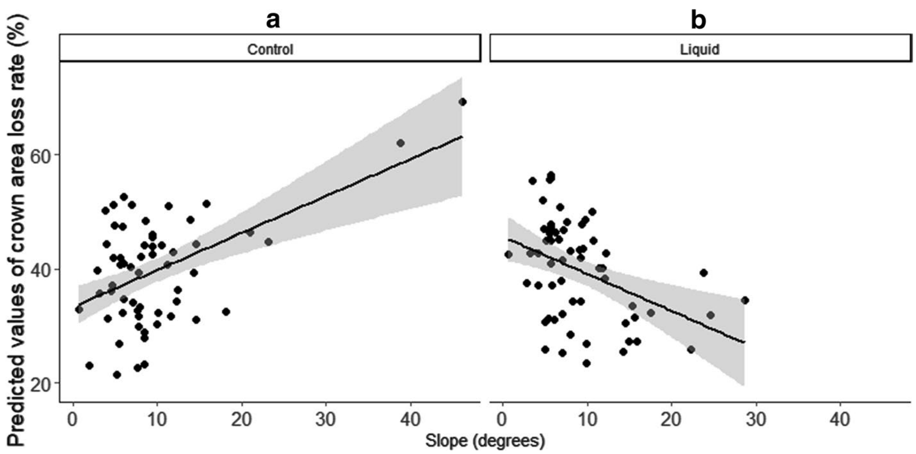

Fig. 3 Predicted values of crown area loss rate (%) depending on treatment-slope interaction (N = 120 trees). In legend, Control = trees within each pair that received no liquid treatment, Liquid = trees within each pair that received a liquid treatment. Different letter above the graphs indicate significant differences ($P < 0.05$)

following decades, both sources of stress are expected to increase within oak-dominated systems due to current global change (Gordon et al. 2004; Peñuelas et al. 2004; IPCC 2007; Collins et al. 2013; Serrano et al. 2017), and therefore new effective practices are needed.

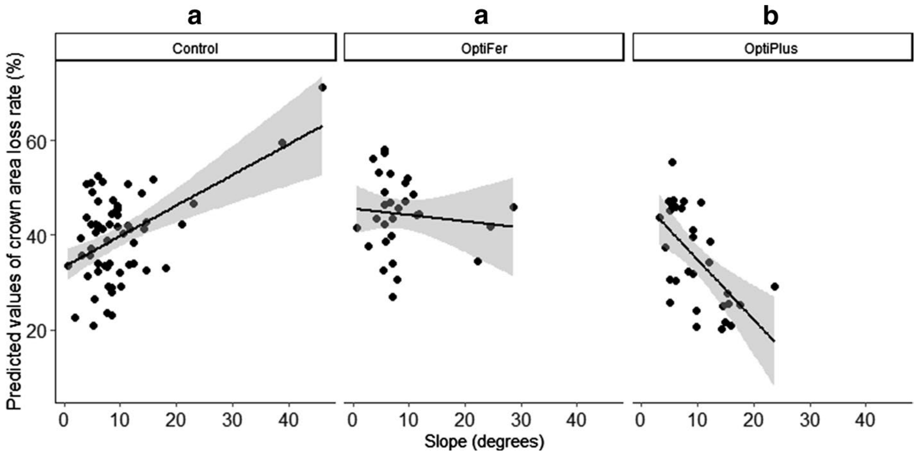

Fig. 4 Predicted values of crown area loss rate (%) depending on product-slope interaction (N = 120 trees). In legend, Control = trees within each pair received no liquid treatment, OptiFer = trees within each pair that received OptiFer liquid treatment, OptiPlus = trees within each pair that received OptiPlus. Different letter above the graphs indicate significant differences ($P < 0.05$)

Table 3 Summary of the top linear mixed models (ΔAIC < 2) to analyze plant herbivory rate (VI models), plant growth rate (VII models) and recruit crown area rate (VIII models) depending on product, oak species and protection

Model	Fixed effects	*Importance*	Levels	*Coeff.*	*SE*	*z-value*	*P*
VI	Intercept			0.028	0.533	0.053	0.958
	Product (P)	1.00	OptiPlus	−0.614	0.578	−1.063	0.288
	Species (S)	0.97	Holm Oak	−1.561	0.714	−2.185	0.029
	Protection (Pr)	1.00	Protected	−2.559	0.488	−5.241	<0.001
	P × S	0.96	OptiPlus × HolmOak	2.421	0.694	3.488	<0.001
	P × Pr	0.99	OptiPlus × Protected	1.562	0.654	2.389	0.017
VII	Intercept			2.101	0.155	13.260	<0.001
	Product (P)	0.40	OptiPlus	0.182	0.215	0.828	0.408
	Species (S)	0.64	Holm Oak	−0.398	0.215	1.807	0.071
VIII	Intercept			1.340	0.140	9.388	<0.001
	Product (P)	0.33	OptiPlus	0.023	0.042	0.540	0.589
	Species (S)	0.37	Holm Oak	0.225	0.265	0.832	0.405

Importance: Importance of predictor variable in the model averaging

Bold type indicates statistical significance ($P < 0.05$)

Results from Product type are against control plants, results for species refer to holm oak against cork oak and results for protection refer to protected plants against non-protected plants

Adult oak trees

One of the visible symptoms of oak decline is the accelerated progressive defoliation (Duque-Lazo et al. 2018; Jung et al. 2018). In this study, we have used two variables to identify the oak decline symptoms in canopies of adult oaks: crown defoliation rate

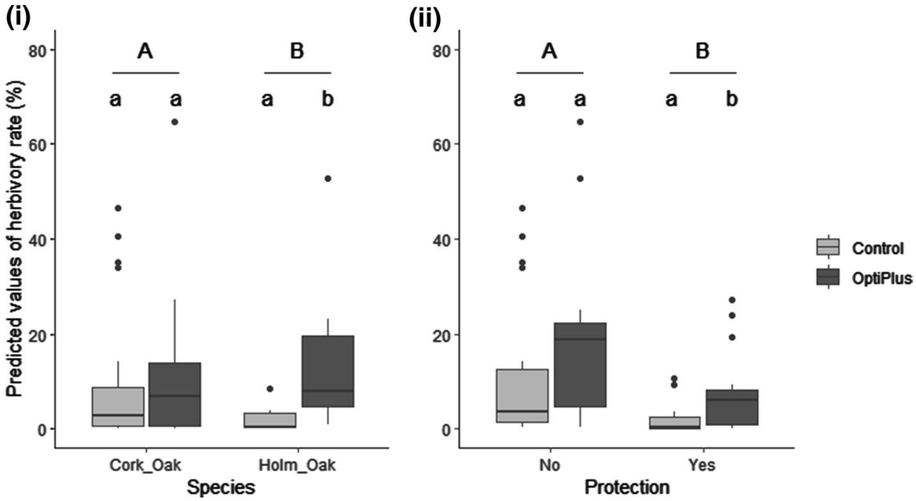

Fig. 5 Predicted values of herbivory rate (%) depending on (i) product-species interaction, and (ii) product-protection interaction (N = 120 recruits). In legend, Control = recruits that received no liquid treatment, OptiPlus = recruits that received OptiPlus treatment. Different letter above the boxes indicate significant differences ($P < 0.05$)

and crown area loss rate. An important issue reported by previous research is that the massive crown defoliation is not only linked to an oomycete presence (e.g. *Phytophthora* spp.); but also to other environmental factors (de Sampaio e Paiva Camilo-Alves et al. 2013). For example, soil characteristics (in particular soil drainage and pH) and water deficit are important drivers of canopy decline (Jung et al. 2018; Sapp et al. 2019). Therefore, the improvement of soil properties through biological fertilizers might be one of the keys to increase oak resilience against *Phytophthora* associated to oak decline. We found a positive effect of OptiPlus product as a soil treatment reducing canopy defoliation; however, for crown area loss rate variable, this amelioriation was only effective in steep slopes. Thus, OptiPlus treatment seemed to buffer the stress generated by an increasing slope, which, in turn, causes shallower and less fertile soils as found in the study area (leptosols) and reduce the water drainage due to higher run-off. Well-drained soils, withsandy to sandy-loamy texture, and acidic (pH < 4) are known to reduce the probability of *Phytophthora* spp. presence (Jung et al. 2000). Therefore, in this study, biological fertilizer such as OptiPlus has increased health status of treated oak trees as we expected (H1).

Regarding OptiFer product, we found a significant positive effect on reducing crown defoliation but it was less effective than OptiPlus since Optifer is a poorer fertilizer, with only three main elements (Fe, Mn, Mg). In contrast, OptiPlus contains greater amount of important deficient minerals [organic N, P, organic P, S, Ca, Mg, and trace elements (Fe, Mn, B, Zn)]. OptiPlus also contains 20% humic fulvic acids, which are important plant-growth stimulators improving nutrient uptake (Kelting 1997; Nikbakht et al. 2008). Particularly, humic concentrations stimulate root development (Kelting et al. 1998), including urban oak trees (Ferrini and Nicese 2002), which may explain a positive response of adult oaks to *Phytophthora*-associated symptoms.

Oak recruits

At oak regeneration level, our results, in general, showed no oak decline symptoms despite they are located in an area severely affected by *Phytophthora* spp. (Sapp et al. 2019). The regeneration was less prone than adult trees to oak decline as it has been also documented in other evergreen Mediterranean *Quercus* affected by *P. ramorum* (Rizzo and Garbelotto 2003; Swiecki and Bernhardt 2006). However, other studies have shown a high susceptibility of holm and cork oak young seedlings to *P. cinnamomi* (Rodríguez-Molina et al. 2002). Therefore, in this study, biological fertilizer (OptiPlus) has not increased growth of treated oak recruits as we expected (H2). For herbivory rate, as expected, we found that wire protectors strongly reduced herbivory on recruits. Many previous studies have reported a herbivory reduction on oak seedlings protected with wire cages (Kochenderfer and Ford 2008; Blossey et al. 2019; López-Sánchez et al. 2019). Importanly, protected recruits that received OptiPlus treatment were more heavily browsed than those protected plants without any treatment. Thus, overall, the fertilized plants were more atractive to herbivores, probably as a result of the greater content in deficient minerals, increasing their palatability (Gambín et al. 2017). Therefore, in this study, treated oak recruits with biological fertilizers (e.g. OptiPlus) have been more heavily browsed as we expected (H3). Hence, it will be necessary to reduce ungulate densities in those areas fertilized in order to reduce herbivory pressure in those treated areas with biological fertilizers.In addition, we found less herbivory damage on holm oaks (*Q. ilex*) than on cork oaks (*Q. suber*) in line with previous research (Bugalho and Milne 2003), which indicates that some species might be more sensitive than others to herbivory damage after treatment.

Recommendations for further research

On the one hand, an important idea pointed out by previous research as well as by this study, is to find adequate tools to improve soil properties (e.g. better management, reduction of soil water deficit and increase in fertility) that minimize oak decline symptoms. On the other hand, herbivory pressure (biotic stress) is expected to increase in the coming decades if the oak-dominated systems are managed inadequately. Therefore further studies should be conducted to analyse the interaction between herbivory and the efficacy of biological fertilizers applied in the soil. In particular, it might be interesting to analyze the application of these products under different types of management (e.g. different ungulate species and densities) and develop soil analysis to examine the possible changes in soil composition (including mycorrhizal communities) after the application of biological fertilizers. In our study area, the general stocking rate was adequate (<0.1 LU ha^{-1}) when compared to other unsustainable systems (López-Sánchez et al. 2016). Adequate grazing management may be a possible tool to improve soil properties along with the application of soil treatments. Soil treatment per se might not work in overgrazed systems as soil compaction may strongly decrease the efficacy of nutrient absorption by the plants (Kozlowski 1999).

Furthermore, the study period was very short; hence, further studies are needed to properly analyze the long-term effects and the mechanisms behind this positive response of trees to fertilizer addition. Hence, we strongly suggest extending the experiment for several years, with multiple times of product application per year (e.g. one in spring and one in autumn) to avoid liquid losses over time and to reinforce the positive effects observed in this preliminary short-term experiment. We also expect that the time of application (e.g. summer vs. winter) will have a significant effect on the response of trees to defoliation as water content and soil compaction strongly vary with time in Mediterranean ungulate-dominated systems.

Finally, it is important to choose the right fertilizer according to soil conditions; especially in poor soils. For example, we found that the fertilizer OptiFer, which contained fewer nutrients and minerals did not apparently cause any significant effect on reducing adult tree defoliation. Therefore, more complete fertilizers, containing a greater variety of deficient nutrients and humic fluvid acids might be more effective, particularly in poor soils.

Implications for management under greater levels of stress

The management intensification exerted in some oak woodlands over the last decades (Pulido et al. 2001; Plieninger et al. 2011; López-Sánchez et al. 2017) contributes to raise the intensity of abiotic and biotic stress agents, particulartly through increased herbivory, soil compaction and pathogen dispersal (Fernández-Rebollo et al. 2009; Metz et al. 2017). Therefore, we strongly recommend avoiding high ungulate densities, particularly in the areas treated with fertilizers to ensure low browsing damage on the fertilized plants. In addition, previous research has suggested cultural actions such as livestock movements to reduce the herbivory pressure in the same areas (Carmona et al. 2013; López-Sánchez et al. 2016; Perea et al. 2016) but with caution in those areas infected by *Phytophthora* spp. (Fernández et al. 2008; Serrano et al. 2017). Here, we applied the fertilizer manually but further studies should consider mechanical fertilization with appropriate machinery around the oaks, ensuring that the deep oak roots reach the nutrients added to the soil. In doing so, early fall seems a suitable time for fertilization, when soil moisture is medium in most Mediterranean systems and when other cultural actions are typically conducted. This practice might be implemented just before acorn drop in areas where acorns are used to feed the domestic or wild ungulates (Fernández-Rebollo et al. 2009).

In sum, the results of this study have slightly visualized the efficacy of some biological fertilizers in reducing *Phythopthora*-associated symptoms and the possibility and need to explore in depth this research line, especially the interaction between herbivory and the efficacy (pros and cons) of using biological fertilizers on oak recruits. Further research efforts should be done not only for conservation and restoration of Mediterranean oak woodlands but also to make them more resilient against global change which is triggering an increase of stress levels.

Acknowledgements We thank Fundación Monte Mediterráneo for the financial support and all facilities to develop the study, especially Ernestine Lüdeke and Hans-Gerd Neglein. We thank Mariana Yuan Ribeiro, Lavinia de Lucchi, Juan de Dios, Johanna Thiede and all Erasmus students from Erasmus + KA1 mobility projects for their help with fieldwork. The project GLOBALFOR supported by UPM and Comunidad de Madrid (Convenio Plurianual for young researchers) also provided financial support.

Author's contribution ALS and RP conceived the ideas and designed the experiments; ALS and RP collected data; ALS analyzed the data; ALS led the writing of the manuscript and RP contributed significantly in the writing of the manuscript.

Funding Fundación Monte Mediterráneo and the project GLOBALFOR supported by UPM and Comunidad de Madrid (Convenio Plurianual for young researchers) provided financial support.

Compliance with ethical standards

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

References

Aukema JE, McCullough DG, Von Holle B, Liebhold AM, Britton K, Frankel SJ (2010) Historical accumulation of nonindigenous forest pests in the continental United States. Bioscience 60:886–897

Barton K (2015) MuMIn: multi-model inference. R Package Version 1(15):1

Bates D, Maechler M, Bolker B, Walker S (2015) Fitting linear mixed-effects models using lme4. J Stat Softw 67:1–48. https://doi.org/10.18637/jss.v067.i01

Blondel J, Aronson J (1999) Biology and wildlife of the Mediterranean region, vol Book. Oxford University Press, Oxford

Blossey B, Curtis P, Boulanger J, Dávalos A (2019) Red oak seedlings as indicators of deer browse pressure: Gauging the outcome of different white-tailed deer management approaches. Ecol Evolut 9:13085–13103

Box GEP, Cox DR (1964) An analysis of transformations. J R Stat Soc Ser B (Methodol) 26:211–252

Branco M, Ramos AP (2012) Coping with pests and diseases. In: Aronson J, Pereira JS, Pausas JG (eds) Cork oak woodlands on the edge: ecology, adaptive management, and restoration. Island Press, Washington, pp 103–113

Brasier CM (1993) Phytophthora cinnamomi as a contributory factor in European oak declines. In: Luisi N, Lerario P, Vannini AB (eds) Recent advances in studies on Oak Decline. Proc. Int. Cong. Selva di Fasano. Tipolitografia-Putignano, Putignano, Italy, pp 49–57

Brasier C, Robredo F, Ferraz J (1993a) Evidence for Phytophthora cinnamomi involvement in Iberian oak decline. Plant Pathol 42:140–145

Brasier CM, Moreira AC, Ferraz JFP, Kirk S (1993b) High mortality of cork oak in Portugal associated with Phytophthora cinnamomi. In: Luisi N, Lerario P, Vannini AB (eds) Recent advances in studies on oak decline. Proc. Int. Cong., Selva di Fasano. Tipolitografia-Putignano, Putignano, Italy, pp 461–462

Broadbent P, Baker KF (1974) Behaviour of Phytophthora cinnamomi in soils suppressive and conducive to root rot. Aust J Agric Res 25:121–137

Bugalho MN, Milne JA (2003) The composition of the diet of red deer (Cervus elaphus) in a Mediterranean environment: a case of summer nutritional constraint? For Ecol Manag 181:23–29

Burney OT, Jacobs DF (2013) Ungulate herbivory of boreal and temperate forest regeneration in relation to seedling mineral nutrition and secondary metabolites. New For 44:753–768

Burnham KP, Anderson DR (2002) Model selection and multi-model inference: a practical information-theoretic approach, 2nd edn. Springer, New York

Campos P, Huntsinger L, Oviedo JL, Starrs PF, Díaz M, Standiford RB, Montero G (2013) Mediterranean oak woodland working landscapes. Landscape Series 16

Caparrós A, Huntsinger L, Oviedo JL, Plieninger T, Campos P (2013) Economics of ecosystem services. In: Campos P et al (eds) Mediterranean oak woodland working landscapes. Springer, Dordrecht, pp 353–388

Carmona CP, Azcárate FM, Oteros-Rozas E, González JA, Peco B (2013) Assessing the effects of seasonal grazing on holm oak regeneration: implications for the conservation of Mediterranean dehesas. Biol Conserv 159:240–247. https://doi.org/10.1016/j.biocon.2012.11.015

Charro JL, López-Sánchez A, Perea R (2018) Traditional cattle vs. introduced deer management in Chaco Serrano woodlands (Argentina): analysis of environmental sustainability at increasing densities. J Environ Manag 206:642–649

Collins M et al (2013) Long-term climate change: projections, commitments and irreversibility. In: Climate change 2013: the physical science basis: contribution of working group I to the fifth assessment report of the intergovernmental panel on climate change. Cambridge University Press, pp 1029–1136

Côté SD, Beguin J, de Bellefeuille S, Champagne E, Thiffault N, Tremblay J-P (2014) Structuring effects of deer in boreal forest ecosystems. Adv Ecol 2014

Dagit R, Carlberg C, Cuba C, Scott T (2015) Economic incentives for oak woodland preservation and conservation. In: Standiford RB, Purcell KL (eds) Proceedings of the seventh California oak symposium: managing oak woodlands in a dynamic world. Gen. Tech. Rep. PSW-GTR-251. U.S. Department of Agriculture, Forest Service, Pacific Southwest Research Station, Berkeley, CA, pp 457–469 de Sampaio e Paiva Camilo-Alves C, da Clara MIE, de Almeida Ribeiro NMC (2013) Decline of Mediterranean oak trees and its association with *Phytophthora cinnamomi:* a review. Eur J Forest Res 132:411–432

Dunne C, Dell B, Hardy GSJ (2003) Biofumigants suppress the growth, sporulation and survival of Phytophthora cinnamomi

Duque-Lazo J, Navarro-Cerrillo RM, Van Gils H, Groen TA (2018) Forecasting oak decline caused by Phytophthora cinnamomi in Andalusia: identification of priority areas for intervention. For Ecol Manag 417:122–136

Duvenhage J, Kotzé J, Maas EM (1992) The influence of nitrogen and calcium on mycelial growth and disease severity of Phytophthora cinnamomi and the effect of calcium on resistance of avocado to root rot. S Afr Avocado Growers' Assoc Yearb 15:12–14

Ellison AM et al (2005) Loss of foundation species: consequences for the structure and dynamics of forested ecosystems. Front Ecol Environ 3:479–486. https://doi.org/10.1890/1540-9295(2005)003%5b0479:lofscf%5d2.0.co;2

Fernández P, Carbonero M, Sánchez M, Trapero A (2008) ¿Cómo manejar las explotaciones afectadas por decaimiento forestal? Europa Cork 36:40–46

Fernández-Olalla M, Martínez-Jauregui M, Perea R, Velamazán M, San Miguel A (2016) Threat or opportunity? Browsing preferences and potential impact of Ammotragus lervia on woody plants of a Mediterranean protected area. J Arid Environ 129:9–15

Fernández-Rebollo P, Sánchez-Hernández M, Serrano-Moral M, García-Moreno A, Carbonero-Muñoz M (2009) La podredumbre radical de encinas y alcornoques en la dehesa

Ferrini F, Nicese F (2002) Response of English oak (*Quercus robur* L.) trees to biostimulants application in the urban environment. J Arboric 28:70–75

Fox J, Weisberg S (2011) An R Companion to Applied Regression. Sage, Thousand Oaks

Gambín P, Ceacero F, Garcia AJ, Landete-Castillejos T, Gallego L (2017) Patterns of antler consumption reveal osteophagia as a natural mineral resource in key periods for red deer (Cervus elaphus). Eur J Wildl Res 63:39

Gentilesca T, Camarero JJ, Colangelo M, Nole A, Ripullone F (2017) Drought-induced oak decline in the western Mediterranean region: an overview on current evidences, mechanisms and management options to improve forest resilience. iFor Biogeosci For 10:796

Gordon IJ, Hester AJ, Festa-Bianchet M (2004) The management of wild large herbivores to meet economic, conservation and environmental objectives. J Appl Ecol 41:1021–1031

Haavik LJ, Billings SA, Guldin JM, Stephen FM (2015) Emergent insects, pathogens and drought shape changing patterns in oak decline in North America and Europe. For Ecol Manag 354:190–205. https://doi.org/10.1016/j.foreco.2015.06.019

Hansen EM (2015) Phytophthora species emerging as pathogens of forest trees. Curr For Rep 1:16–24. https://doi.org/10.1007/s40725-015-0007-7

Heyman F, Lindahl B, Persson L, Wikström M, Stenlid J (2007) Calcium concentrations of soil affect suppressiveness against Aphanomyces root rot of pea. Soil Biol Biochem 39:2222–2229

Journée d'infomation sur la Réserve de Biosphère des Oasis de Sud Marocain. In: Journée d'infomation sur la Réserve de Biosphère des Oasis de Sud Marocain, 2007

IPCC (2007) Climate change 2007: synthesis report. An assessment of the intergovernmental panel on climate change. IPCC, Cambridge

IPCC (2014) Climate Change 2014: Synthesis report. Contribution of working groups I, II and III to the fifth assessment report of the intergovernmental panel on climate change. Geneva, Switzerland

Johnstone JF et al (2016) Changing disturbance regimes, ecological memory, and forest resilience. Front Ecol Environ 14:369–378

Jones JD, Dangl JL (2006) The plant immune system nature. Nature 444:323–329

Jung T, Blaschke H, Oßwald W (2000) Involvement of soilborne Phytophthora species in Central European oak decline and the effect of site factors on the disease. Plant Pathol 49:706–718

Jung T, Pérez-Sierra A, Durán A, Jung MH, Balci Y, Scanu B (2018) Canker and decline diseases caused by soil-and airborne Phytophthora species in forests and woodlands. Persoonia Mol Phylogeny Evol Fungi 40:182

Kelting MP (1997) Effects of soil amendments and biostimulants on the post-transplant growth of landscape trees. Virginia Tech

Kelting M, Harris JR, Fanelli J, Appleton B (1998) Humate-based biostimulants affect early post-transplant root growth and sapflow of balled and burlapped red maple. HortScience 33:342–344

Kochenderfer JN, Ford WM (2008) Utility of wire cages, tree shelters, and repellants to minimize herbivory to oak by white-tailed deer Res Pap NRS-5 Newtown Square, PA: US Department of Agriculture, Forest Service, Northern Research Station 8 p 5

Kozlowski T (1999) Soil compaction and growth of woody plants. Scand J For Res 14:596–619
López-Sánchez A (2015) Balancing management and preservation of Mediterranean scattered oak woodlands (dehesas) in human-dominated landscapes. Universidad Politécnica de Madrid (Spain)
López-Sánchez A, Schroeder J, Roig S, Sobral M, Dirzo R (2014) Effects of cattle management on oak regeneration in northern Californian Mediterranean oak woodlands. PLoS ONE 9:e105472. https://doi.org/10.1371/journal.pone.0105472
López-Sánchez A, Perea R, Dirzo R, Roig S (2016) Livestock vs. wild ungulate management in the conservation of Mediterranean Dehesas: implications for oak regeneration. For Ecol Manag 362:99–106. https://doi.org/10.1016/j.foreco.2015.12.002
López-Sánchez A, Dirzo R, Roig S (2017) Changes in livestock footprint and tree layer coverage in Mediterranean dehesas: a six-decade study based on remote sensing. Int J Remote Sens 39:4727–4743
López-Sánchez A, Peláez M, Dirzo R, Fernandes GW, Seminatore M, Perea R (2019) Spatio-temporal variation of biotic and abiotic stress agents determines seedling survival in assisted oak regeneration. J Appl Ecol 00:1–12
Lutz JA, Van Wagtendonk JW, Franklin JF (2010) Climatic water deficit, tree species ranges, and climate change in Yosemite National Park. J Biogeogr 37:936–950
Metz MR, Varner JM, Simler AB, Frangioso KM, Rizzo DM (2017) Implications of sudden oak death for wildland fire management. For Phytophthoras 7:30–44
Miranda M, Díaz L, Sicilia M, Cristóbal I, Cassinello J (2011) Seasonality and edge effect determine herbivory risk according to different plant association models. Plant Biol 13:160–168
Møller AP, Shykoff JA (1999) Morphological developmental stability in plants: patterns and causes. Int J Plant Sci 160:S135–S146
Müller E, Stierlin HR (1990) Sanasilva: Kronenbilder: mit Nadel-und Blattverlustprozenten. Eidgenössische Forschungsanstalt für Wald, Schnee und Landschaft, Birmensdorf, Switzerland
Navarro RM, Ariza D, Porras C, Jorge I, Jorrín J (2009) Evaluación de la resistencia aparente de individuos de encina a Phytophthora cinnamomi Rands. Boletín de sanidad vegetal Plagas 35:89–97
Nikbakht A, Kafi M, Babalar M, Xia YP, Luo A, Etemadi N-A (2008) Effect of humic acid on plant growth, nutrient uptake, and postharvest life of gerbera. J Plant Nutr 31:2155–2167
Nolte DL (2003) Managing ungulates to protect trees USDA National Wildlife Research Center-Staff Publications 261
Nolte DL, Dykzeul M (2000) Wildlife impacts on forest resources. Human conflicts with wildlife: economic considerations 20
Oh JH, Jones MB, Longhurst WM, Connolly GE (1970) Deer browsing and rumen microbial fermentation of Douglas-fir as affected by fertilization and growth stage. For Sci 16:21–27
Peñuelas J et al (2004) Nonintrusive field experiments show different plant responses to warming and drought among sites, seasons, and species in a north–south European gradient. Ecosystems 7:598–612
Perea R, Gil L (2014) Shrubs facilitating seedling performance in ungulate-dominated systems: biotic versus abiotic mechanisms of plant facilitation. Eur J For Res 133:525–534
Perea R, Girardello M, San Miguel A (2014) Big game or big loss? High deer densities are threatening woody plant diversity and vegetation dynamics. Biodivers Conserv 23:1303–1318
Perea R, Perea-García-Calvo R, Díaz-Ambrona CG, San Miguel A (2015) The reintroduction of a flagship ungulate Capra pyrenaica: assessing sustainability by surveying woody vegetation. Biol Conserv 181:9–17
Perea R, López-Sánchez A, Roig S (2016) The use of shrub cover to preserve Mediterranean oak dehesas: a comparison between sheep, cattle and wild ungulate management. Appl Veg Sci 19:244–253
Perea R, López-Sánchez A, Pallarés J, Gordaliza GG, González-Doncel I, Gil L, Rodríguez-Calcerrada J (2020) Tree recruitment in a drought- and herbivory-stressed oak-beech forest: Implications for future species coexistence. For Ecol Manag 477:118489
Plieninger T, Wilbrand C (2001) Land use, biodiversity conservation, and rural development in the dehesas of Cuatro Lugares. Spain Agrofor Syst 51:23–34
Plieninger T, Pulido FJ, Konold W (2003) Effects of land-use history on size structure of holm oak stands in Spanish dehesas: implications for conservation and restoration. Environ Conserv 30:61–70
Plieninger T, Schaich H, Kizos T (2011) Land-use legacies in the forest structure of silvopastoral oak woodlands in the Eastern Mediterranean. Reg Environ Change 11:603–615
Provenza FD, Villalba JJ, Dziba L, Atwood SB, Banner RE (2003) Linking herbivore experience, varied diets, and plant biochemical diversity. Small Ruminant Res 49:257–274
Pulido FJ, Díaz M (2005) Regeneration of a Mediterranean oak: a whole-cycle approach. Ecoscience 12:92–102

Pulido FJ, Díaz M, de Trucios SJH (2001) Size structure and regeneration of Spanish holm oak Quercus ilex forests and dehesas: effects of agroforestry use on their long-term sustainability. For Ecol Manag 146:1–13
R Development Core Team (2019) R: a language and enviroment for statistical computing, 3.6.0 edn. Foundation for Statistical Computing, Vienna, Austria
Ríos P, Obregón S, de Haro A, Fernández-Rebollo P, Serrano MS, Sánchez ME (2016) Effect of Brassica biofumigant amendments on different stages of the life cycle of Phytophthora cinnamomi. J Phytopathol 164:582–594
Rivas-Martínez S, Fernández-González F, Loidi J, Lousã M, Penas A (2001) Syntaxonomical checklist of vascular plant communities of Spain and Portugal to association level vol 14. Itinera geobotanica,
Rizzo DM, Garbelotto M (2003) Sudden oak death: endangering California and Oregon forest ecosystems. Front Ecol Environ 1:197–204
Rodríguez-Molina M, Torres-Vila L, Blanco-Santos A, Nunez EP, Torres-Álvarez E (2002) Viability of holm and cork oak seedlings from acorns sown in soils naturally infected with Phytophthora cinnamomi. For Pathol 32:365–372
Ruiz-Gómez FJ, Navarro-Cerrillo RM, Pérez-de-Luque A, Oβwald W, Vannini A, Morales-Rodríguez C (2019) Assessment of functional and structural changes of soil fungal and oomycete communities in holm oak declined dehesas through metabarcoding analysis. Sci Rep 9:1–16
San Miguel A, Perea R, Fernández-Olalla M (2010) Wild ungulates vs extensive livestock. Looking back to face the future. Opt Méditerr A 92:27–34
Sánchez M, Caetano P, Ferraz J, Trapero A (2002) Phytophthora disease of Quercus ilex in south-western Spain. For Pathol 32:5–18
Sapp M et al (2019) Site-specific distribution of oak rhizosphere-associated oomycetes revealed by cytochrome c oxidase subunit II metabarcoding. Ecol Evol 9:10567–10581
Serrano M, De Vita P, Carbonero M, Fernández F, Fernández-Rebollo P, Sánchez M (2012a) Susceptibility to Phytophthora cinnamomi of the commonest morphotypes of holm oak in southern Spain. For Pathol 42:345–347
Serrano MS, De Vita P, Fernández-Rebollo P, Hernández MES (2012b) Calcium fertilizers induce soil suppressiveness to Phytophthora cinnamomi root rot of Quercus ilex. Eur J Plant Pathol 132:271–279
Serrano MS, Ríos P, González M, Romero MÁ, Fernández P, Sánchez ME (2017) A review of integrated control of Phytophthora root rot in oak rangeland ecosystems IOBC-WPRS. Bulletin 127:139–146
Swiecki TJ, Bernhardt EA (2006) A field guide to insects and diseases of California oaks Gen Tech Rep PSW-GTR-197 Albany, CA: Pacific Southwest Research Station, Forest Service, US Department of Agriculture 151 p 197
Tyler CM, Kuhn B, Davis FW (2006) Demography and recruitment limitations of three oak species in California. Q Rev Biol 81:127–152
Underwood EC, Viers JH, Klausmeyer KR, Cox RL, Shaw MR (2009) Threats and biodiversity in the mediterranean biome. Divers Distrib 15:188–197
Venables WN, Ripley BD (2002) Modern applied statistics with S, vol Book. Springer, Berlin
White PS, Jentsch A (2001) The search for generality in studies of disturbance and ecosystem dynamics. In: Esser K, Lüttge U, Kadereit JW, Beyschlag W (eds) Progress in Botany, vol 62. Progress in Botany. Springer, Berlin, pp 399–450. https://doi.org/10.1007/978-3-642-56849-7_17
Yang C, Liang Y, Qiu D, Zeng H, Yuan J, Yang X (2018) Lignin metabolism involves Botrytis cinerea BcGs1-induced defense response in tomato. BMC Plant Biol 18:103
Zamora R, Gómez JM, Hódar JA, Castro J, García D (2001) Effect of browsing by ungulates on sapling growth of Scots pine in a Mediterranean environment: consequences for forest regeneration. For Ecol Manag 144:33–42

Affiliations

Aida López-Sánchez[1] · **Ramón Perea**[1]

✉ Aida López-Sánchez
aida.lopez@upm.es

[1] Departamento de Sistemas y Recursos Naturales, Universidad Politécnica de Madrid, Ciudad Universitaria, s/n, 28040 Madrid, Spain