# Crop production, recruitment and survival in *Quercus ilex*: density-dependent effects in a mast-seeding species

*Holm oak seedling*
*Photograph by Mario Díaz*

MÁSTER UNIVERSITARIO EN RESTAURACIÓN DE ECOSISTEMAS

TRABAJO FIN DE MÁSTER


**Presentado por:**

María Teresa Sánchez Mejía

**Director:**

Dr. Mario Díaz Esteban

**Tutor académico:**

Dra. María Dolores Jiménez Escobar


**Madrid, Junio de 2020**

TABLE OF CONTENT

Abstract ..... 3
Keywords ..... 3
Resumen ..... 4
Palabras clave ..... 4
1. Introduction ..... 5
1.1. The relevance of natural regeneration and masting ..... 5
1.2. Drivers of masting ..... 6
1.3. Density-dependence dynamics in a mast species ..... 7
1.4. Masting and density-dependence in *Quercus ilex* ..... 9
2. Objectives and hypothesis ..... 10
3. Methodology ..... 11
3.1. Study species ..... 11
3.2. Study area ..... 12
3.3. Study design and survey of crops and saplings ..... 13
3.4. Climatic variables ..... 17
3.5. Statistical Analysis ..... 17
4. Results ..... 20
4.1. First-year recruitment ..... 22
4.2. Long-term survival ..... 23
4.3. Seedling populations ..... 28
5. Discussion ..... 29
*5.1.* Crop production and masting in *Quercus ilex* ..... 29
5.2. Effects of masting and seedling density on first-year recruitment ..... 30
5.3. Effects of masting and density in seedling survival ..... 32
5.4. Effects of tree cover and climate ..... 34
5.5. Seedling fate and evolution of masting ..... 34
5.6. Limitations of the study ..... 36
6. Conclusions ..... 37
7. Agradecimientos ..... 39
8. Literature cited ..... 39
APPENDIX 1. DETERMINATION OF MAST YEARS ..... 46
APPENDIX 2. SPEIs (DROUGHT INDEX) ..... 47

## LIST OF FIGURES

***Figure 1.*** Density-dependent effects in a mast-seeding species……..... 8

***Figure 2.*** Reproductive cycle of the Holm oak (*Quercus ilex* subsp. *ballota*)……..... 10

***Figure 3.*** Climodiograms of the studied locations ……..……..... 12

***Figure 4.*** Location of the study……..... 14

***Figure 5.*** a) Acorns on Holm Oak branches..……...…..... 15

b) Tagged newly emerged seedlings

c) Delimited transect by a holm oak.

***Figure 6.*** Seedling and acorn production dynamics……..... 21

***Figure 7***. Interaction plots of the GLM Negative Binomial Models (emergence)……..... 25

a) Acorn crop:Location

b) Seedling density (t+1):Location

c) Acorn crop:SPEI9_FO(t+1)

d) Acorn crop:Tree cover; e)Acorn crop: Seedling density(t+1)

f) Tree cover: Seedling density(t+1)

g) SPEI9_FO:Acorn crop:Location

h) Location:Acorn crop: Seedling density (t+1);

i) Acorn crop: Seedling density (t+1): SPEI9_FO (t+1).

***Figure 8.*** Interaction plots of the Cox hazard model (survival)……..……..……. 26

a) Seedling density: Cohort

b) Seedling density: SPEI3_V

c) Seedling density:Tree cover

d) Cohort:Seedling density:Tree cover

e) Tree cover:Seedling density: Cohort.

***Figure 9.*** Kaplan Meier Survival curves ……..…….……..... 27

***Figure 10***. Seedling bank dynamic of the studied locations…….……..…..….... 28

***Figure 11.*** Distribution of Quercus ilex in Montejo de la Vega……..……. 30

***Figure 12***. Seedlings in a cache…… 32

## LIST OF TABLES

***Table 1.*** Location and information of the sites included in the study…….. 13

***Table 2.*** Census data of holm oak emergence in the two areas of study ……. 20

***Table 3.*** Summary of GLMM (Negative Binomial) and COX mixed-model…….. 24

## Abstract

Masting, the synchronous and irregular production of large crops over time, is adaptive if improved short-term recruitment during mast years was not compensated by increased density-dependent seedling mortality in the subsequent years. However, only a handful of studies have addressed the relationships between masting, seedling density, and long-term recruitment needed to understand the underlying advantage of intermittent large crop production.

Acorn crops, first-year recruitment, and long-term survival of Holm oak (*Q. ilex* subsp. *ballota)* seedlings were monitored in two contrasting study locations during 1998-2019 and 2001-2019. Relationship between variable crops, early recruitment, seedling survival and conspecific seedling density were analyzed, together with the modulating effects of local tree cover and climate. First-year recruitment was analyzed by fitting generalized linear models, while survival curves and hazard models were used to analyze factors affecting long-term survival.

Crop production enhanced first-year recruitment but increased the risk of seedling mortality in the long-term, especially in the location where oaks showed a more typical masting behavior. Seedling density showed positive effects associated with the aggregation of seedlings in safe sites, however, in the long-term, when conditions were favorable for seedling establishment, density had a negative effect related to higher competition and natural enemies attraction. Besides, tree cover and climate interacted with the effects of masting and density by ameliorating the stressful environmental conditions and reducing intraspecific competition. As a consequence, great differences between locations were found, with higher mortality associated with density-dependence in the most prone to masting, and greater limitation by safe sites where mast events are softer. The results obtained show the importance of long-term approaches to understand the contribution of masting and density-dependence to early recruitment, as well as the importance of including local factors to avoid masking relevant effects.

**Keywords**: Masting, Early recruitment, Density-dependence, Seedling dynamics, Holm oak, Local factors.

## Resumen

La vecería, la producción sincrónica e irregular de grandes cosechas, es adaptativa si el incremento en el reclutamiento temprano en los años veceros no es compensado por una mayor mortalidad asociada a factores denso-dependendientes. Sin embargo, solo unos pocos estudios a largo plazo han abordado la relación entre vecería, densidad de plántulas y reclutamiento necesarios para entender la ventaja evolutiva de la producción intermitente de grandes cosechas.

La producción de cosecha, el reclutamiento durante el primer año y la supervivencia a largo plazo de plántulas de encina (*Quercus ilex* subsp. *ballota*) fue monitoreado en dos localidades contrastadas durante 1998-2019 y 2001-2019. La relación entre variabilidad de la cosecha, reclutamiento temprano, supervivencia de plántulas y densidad intraespecífica han sido analizados junto a los efectos moduladores de la cubierta arbórea y el clima. El reclutamiento en el primer año se analizó mediante modelos lineales generalizados, mientras que para la supervivencia se emplearon curvas de supervivencia y modelos de riesgo.

La vecería tuvo un efecto positivo inicial, pero mayor mortalidad a largo plazo, especialmente en la localidad más vecera. La densidad mostró efectos positivos asociados a la agregación de plántulas en sitios favorables, pero a largo plazo, en condiciones favorables para las plántulas tuvo un efecto negativo ligado a la competencia y a atracción de enemigos naturales. Además, la cobertura y el clima interactuaron con la vecería y la densidad suavizando las condiciones ambientales estresantes y la competencia intraespecífica. En consecuencia, se observaron grandes diferencias entre localidades, con mayor mortalidad asociada a mecanismos denso-dependientes en la zona más vecera y mayor limitación por sitios favorables en la zona menos vecera. Los resultados obtenidos muestran la importancia de enfoques a largo plazo para comprender la contribución de la vecería y denso-dependencia en el reclutamiento temprano, así como la importancia de la inclusión de factores locales.

**Palabras clave**: Vecería, Reclutamiento temprano, Denso-dependencia, Dinámica de plántulas, Encina, Factores locales.

# 1. Introduction

## 1.1. The relevance of natural regeneration and masting

Forests in almost every bioregion of the world are suffering severe losses in biodiversity, dynamics, and functioning (Keenan, et al., 2015). Hence, maintaining the capacity of forests to regenerate naturally has become a crucial question, and a conservation goal of current management policies (Chazdon & Uriarte, 2016).

Natural regeneration is a multistage process that ensures the long-term persistence of a plant population by the replacement of the old mature individuals by the new recruits. Regeneration is strongly determined by the processes that occurred in the early stages of seed production, dispersal, successful seed germination, and seedling survival until recruitment (Johnson, et al., 2014). Outcomes of these early-stage processes have however far-reaching impacts on the structure and dynamics of forest communities (Harper, 1977).

Most species present fluctuations in seed production over the years, but some plants extreme this behavior by maximizing their reproductive output (flowers and seeds) in some years (*mast* years), followed by subsequent years with little reproductive activity. This strategy is known as masting or mast-seeding, and is characterized by high interannual variability of seed crops with high synchronicity at the population level over large areas (Janzen, 1974; Sork, et al., 1993; Kelly, 1994; Kelly & Sork, 2002). Masting is taxonomically and geographically widespread. It seems to be more prevalent in temperate ecosystems, mainly in the Fagaceae and Pinaceae families (Sork, et al., 1993; Kelly & Sork, 2002; Koening & Knops, 2013), but there are also examples in tropical Dipterocarpaceae, Podocarpaceae and Fabaceae species (Janzen, 1974; Henkel, et al., 2005; Visser, et al., 2011; Oshima, et al., 2015; Henkel & Mayor, 2019).

The extreme variations in the reproductive output of mast species have direct consequences in their regeneration, as usually found when comparing seed predation and dispersal and early seedling recruitment between masts and non-mast years (Li & Zhang, 2007; Espelta, et al., 2009; Joshi & Tewari, 2009; Archibald, et al., 2012; Xiao, et al., 2013; Henkel & Mayor, 2019), but also in the whole ecosystem through bottom-up effects. The ephemeral pulse of seed resources may regulate the population of consumers and pathogens, which may involve larger

effects at trophic levels (Soler, et al., 2017; Bogdziewicz, et al., 2018; Masaki, et al., 2020), even including disease outbreaks (Ostfeld & Keesing, 2000; Tersago, et al., 2009). Due to the significant implications in ecosystem composition and dynamics, and forest regeneration; understanding masting and its effects is crucial to assess ecosystem resilience and adaptation and thereby it will improve our ability to design conservation and management strategies (Kettle, 2012; Masaki, et al., 2020). No-studies, however, have addressed whether these early-stage effects of masting remains until effective recruitment to the adult population.

### 1.2. Drivers of masting

Masting is characterized by two mechanistic processes occurring at the individual and the population level, respectively: interannual variability and synchronicity (Kelly, 1994). How this synchronicity and variability occur have been explained through two-sets of non-exclusive hypotheses, proximate-level mechanisms on a short-term scale, and evolutionary reproductive strategies on a long-term scale (Kelly, 1994; Pearse, et al., 2016), with several studies suggesting that the identity and effect of these hypotheses may vary among species and ecosystems, and masting could be triggered by a combination of mechanisms (Kelly & Sork, 2002; Pearse, et al., 2016).

Environmental factors, pollen availability and coupling, and resource allocation strategies have been identified as important proximate drivers of masting, although these mechanisms are not necessarily based on the adaptive consequences of masting from an evolutionary point of view (Pearse, et al., 2016). Two of the most classical and supported proximate mechanisms driving masting are the Moran effect, that explains synchronic crop production among individuals based on spatial patterns of synchrony in weather conditions (Koening & Knops, 2005); and de Resource Budget Model, that posits that reproduction requires the storage of resources for several years until a certain threshold is reached because masting completely depletes such stores (Pearse, et al., 2016). This last hypothesis has been also postulated to explain the prevalence of masting in less productive environments, like the Mediterranean basin (Crone, et al., 2011).

The evolutionary advantages of masting have been hypothesizing to happen trough one or more non-exclusive economies of scale, with two hypotheses with the most empirical support, The Greater Pollination Efficiency and The Predator Satiation (Norton & Kelly, 1988; Sork, et al., 1993; Kelly, 1994; Pearse, et al., 2016). Synchronized flowering effort in some years will increase pollination success resulting in larger crop production (Kelly, et al., 2001; García-Mozo, et al., 2007; Moreira, et al., 2014; Meyer & Pendleton, 2015). On the other hand, masting will enhance seed scape because of food-shortage during non-mast years reduce predators populations that are unable to consume the much larger crops during mast years (Janzen, 1974; Visser, et al., 2011; Meyer & Pendleton, 2015; Moreira, et al., 2016; Xiao, et al., 2017; Bogdziewicz, et al., 2018). In any case, despite the differences among the evolutionary hypothesis of masting, all of them are based on the assumption made by Norton & Kelly (1988) of "large reproductive episodes are more efficient than small ones"; and thereby in mast species, the high density of conspecific (flowers, seeds or seedlings) should have a positive effect on recruitment.

### 1.3. Density-dependence dynamics in a mast species

Apart from advantages, masting imposes clear disadvantages for both parental trees and offspring, including missing reproductive opportunities or lower growth rates in trees (Waller, 1979; Norton & Kelly, 1988; Kelly, 1994; Martín, et al., 2015; Hacket-Pain, et al., 2017), lower seed dispersal by satiation of dispersers (Herrera, et al., 1994) and high mortality of the seedlings after emergence (Hett, 1971). Therefore, masting can only be a viable evolutionary strategy when the benefits compensate for costs (Visser, et al., 2011; Oshima, et al., 2015). Furthermore, whether the advantage of greater reproductive efficiency at high reproductive effort (Norton & Kelly, 1988) influences the evolution of masting is strongly dependent on the processes occurring in later developmental stages (Allen, et al., 2017).

Seedling-stage is a crucial bottleneck for plant regeneration (Pulido & Díaz, 2005; Queenborough, et al., 2007; Martini, et al., 2019). The high vulnerability of seedlings to natural enemies and abiotic stresses affect their performance, but also, the density and composition of neighboring individuals, especially when they belong to the same species (Comita, et al., 2014;

Johnson, et al., 2014; Zhu, et al., 2015). High local conspecific densities, as occurs after a mast event, have been associated with a negative impact in the survival, growth, or recruitment of individuals by increasing the intraspecific competition or the attraction of specialist pathogens and predators (Janzen, 1970). Strong evidence of this conspecific negative density-dependent (CNND) effects have been addressed in several community-level studies in temperate forest (Piao, et al., 2013; Johnson, et al., 2014; LaManna, et al., 2016) and, most commonly, in tropical forests (Connell & Green, 2000; Oshima, et al., 2015; Calderón-Sanou, et al., 2019; Martini, et al., 2019), where the CNND model has been frequently proposed as a promoter of forest biodiversity (Comita, et al., 2014).

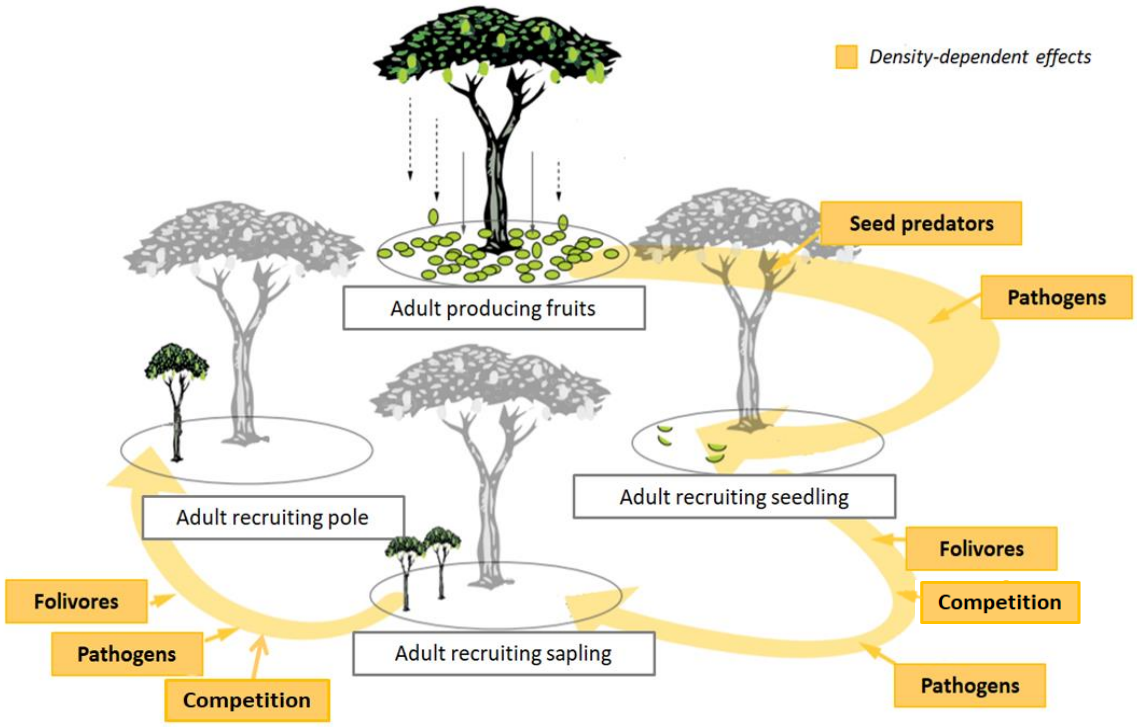


***Figure 1***. Density-dependent effects in a mast-seeding species

Modified from Beaune et al. (2012)

Consequently, the positive density-dependent effects attributed to masting could be attenuated by negative-density dependent effects related to competition and attraction of natural enemies (Comita, et al., 2014), with some authors suggesting a prevalence of positive effects of masting before seed dispersal and negative effects in later stages (Oshima, et al., 2015; Xiao, et al., 2017; Bogdziewicz, et al., 2018). So far, just a bunch of studies, frequently short-term (3 years

at most), have analyzed the density-dependent dynamics of seedlings in mast species. Mixed results have been reported, ranging from prevalence of negative density-dependent effects over survival (Oshima, et al., 2015), positive effects (Calderón-Sanou, et al., 2019), mixed-effects (Connell & Green, 2000; Lin, et al., 2012), and frequently not showing any clear effect at all (Martini, et al., 2019).

Apart from logistic constraints, scarcity of long-term studies relating masting to effective seedling recruitment are partly due to the assumption that processes acting during the earlier stages of seed production, dispersal, germination, and early establishment play a crucial role in plant regeneration (Herrera, et al., 1994; Marañón 2008; Pulido, et al., 2010). However, final recruitment requires the survival of seedlings for many subsequent years to reach incorporation to the reproductive population (Frey, et al., 2007) in which it could be regulated by the effects of mast-seeding on density-dependence. Further long-term survival and density-dependent effects are expected to be modulated by climatic variation at regional scales, by local variation of key factors such as light availability, nutrient sources or herbivory, and by intrinsic factors of seedlings such as age or size. The relative impact, direction, and interactive effects of these factors are susceptible to temporal variation (Frey, et al., 2007). Thereby the contribution of masting to effective recruitment and therefore its evolutionary meaning as either an adaptive strategy or a by-product of proximate mechanism, requires long-term approaches to be fully addressed.

### 1.4. Masting and density-dependence in *Quercus ilex*

Most studies addressing density dependence in mast species have been carried out in tropical environments (Connell & Green, 2000; Oshima, et al., 2015; Calderón-Sanou, et al., 2019; Martini, et al., 2019) although previous studies in temperate environments have shown that density could be playing an important role in early recruitment (Piao, et al., 2013; Johnson, et al., 2014; LaManna, et al., 2016). Holm oaks (*Quercus ilex*) can be adequate for addressing the effects of masting and density-dependence mechanisms in early recruitment and seedling survival, as it has been extensively studied in the last decades.

Holm oaks are widespread in the Mediterranean basin, being the dominant species in most forests of the Iberian Peninsula (Blanco, et al., 1997). Despite masting has been widely studied

in this species, the focus has been mostly on the drivers of masting (García-Mozo, et al., 2007; Pérez-Ramos, et al., 2010; Koenig, 2013), or the effect of masting on predispersal stages (Espelta, et al., 2008; Bogdziewicz, et al., 2018). A handful of papers have also explored effects of forest management on acorn production, predispersal losses and acorn dispersal (Marañón, 2008; Díaz & Pulido, 2009; Díaz, et al., 2011; Díaz, 2014; Valladares, et al., 2014). Nevertheless, no long-term study on seedling dynamics is still available for this species, a fact that questions the adaptive interpretations of masting patterns found to date.

## 2. Objectives and hypothesis

The main purpose of this long-term study is to examine the effect of crop production over early recruitment associated with the phenomenon of masting in holm oaks, as well as the change of recruitment patterns over time and the effect of denso-dependence mechanism on compensating the initial patterns of masting.

The following hypotheses were tested:

- (H1) Density-dependence effects may partially outweigh the effects of masting in seedling recruitment.
- (H2) The main factors involved in seedling recruitment may change over time, with an initial prevalence of crop production and the latter dominance of density-dependence mechanisms or local factors.
- (H3) Local factors may play an important role in the effects of masting and density-dependence over recruitment.
- (H4) Mast years contribute largely to the seedling population than non-mast years.

For this purpose, long-term monitoring of crop production and seedling dynamics of *Quercus ilex* subsp. *ballota* has been carried out in two contrasting locations of the Iberian Peninsula, from 1998 to 2019 in Navas de Estena (Ciudad Real) and 2001-2019 in Montejo de la Sierra (Segovia). Specifically, the study has been focused on the effects of masting, density-dependence, and local factors over first-year recruitment and long-term survival.

# 3. Methodology

## 3.1. Study species

The holm oak (*Quercus ilex* subsp. *ballota)* is as a late-successional species that shows a typical mast behavior (Koenig et al. 2013). Wind pollination occurs between April and May, fruit development during summer and early autumn, and acorn fall between October and January (*Fig.* 1; Vázquez, 1997). A variable proportion of acorns is lost to predispersal insect seed predators in the genus *Curculio* and *Cydia*, mainly (Díaz, et al., 2011; Díaz, 2014). Fallen acorns remain on the surface exposed to predation and dispersals by vertebrates (mainly deer *Cervus elaphus* and *Capreolus capreolus*, wild boar *Sus scrofa*, mice *Apodemus sylvaticus* and *Mus spretus* and European jays *Garrulus glandarius*; Díaz et al. 2011, Díaz 2014). Seedling germination occurs in winter and the emergence of seedlings between March and June (Pulido & Díaz, 2005).

***Figure 2.*** Reproductive cycle of the Holm oak (*Quercus ilex* subsp. *ballota*)

Numbers indicate the phases of the reproductive cycle (1: flowering; 2: fertilization; 3: growth / abortion of fruits, 4: viable fruit ripening; 5: dispersal; 6: emergence: 7: seedling establishment, and 8: growth), Outer part are represented the main factors involved in reproductive loses (insect herbivory, predispersal, dispersal, and postdisperal seed predation, and seedling and sapling losses due to drought or herbivory).

Source: Díaz et al. (2011).

### 3.2. Study area

The study was carried out in two different protected areas of Spain (*Fig.* 4), the Natural Park of the Hoces del Río Riaza (Montejo de la Vega, Segovia), and the National Park of Cabañeros (Navas de Estena, Ciudad RealThe climate in both, Montejo (3°35'10.16"W, 41°32'20.51"N) and Estena (4°32'54.84"W, 39°30'10.15"N), is the typical Mediterranean, with mild winters, dry and warm summers, and rainfall concentrated in spring and autumn. According to the Köppen climate classification, Montejo and Estena correspond to Csb and Csa climate, respectively; thus Estena is characterized by greater aridity in summer (*Fig. 3* and Table 1). The locations were chosen as they represent two contrasting areas for *Quercus ilex* subsp. *ballota*, with more favorable climatic conditions in Estena, whereas the populations of Montejo are on the edge of its distribution, being substituted by *Juniperus* spp. in the southern slopes.

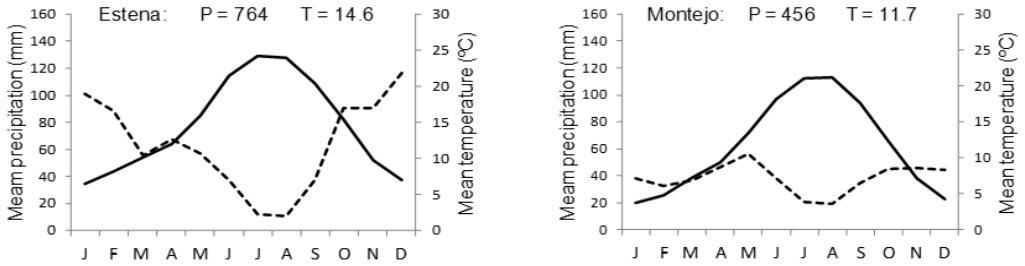


***Figure 3.*** Climodiograms of the studied locations (left: Estena; right: Montejo).

The continuous line represents mean Temperature (ºC) and dotted-line mean precipitation (mm). Mean long-term data from the official AEMET stations of Navas de Estena (1997-2010; n=13 years) and Linares de los Arroyos (1962-2019; n=57 years), respectively, available from nearby stations.

In both locations, the plant community is largely dominated by a fairly continuous overstory of *Quercus ilex* subsp. *ballota* (holm oak), with scattered Lusitanian *Q. faginea* oaks and Spanish junipers *Juniperus thurifera* in Montejo, and Lusitanian, cork *Q. suber* and Pyrenean *Q. pyrenaica* oaks in Navas de Estena. The main understory shrubs are *Amelanchier ovalis*, *Genista scorpius*, and several species of the genus *Thymus*, *Lavandula*, *Dorycnum*, and *Lonicera* in Montejo, and *Phillyrea angustifolia* L., *Pistacia terebinthus* L., *Cistus ladanifer* L. and *Arbutus unedo* L. in Navas de Estena. Current management consists of occasional understory clearance to prevent wildfires, maintenance on paths for recreative visits, and very

occasional extensive grazing and big game hunting. Past management (until the 50's-60's of the XX century) included extensive grazing, firewood collection and charcoal production.

***Table 1.*** *Location and information of the sites included in the study*

| | Montejo de la Vega | Navas de Estena |
|---|---|---|
| Altitude (m) | 900-930 | 600-800 |
| Substrate | Calcareous (limestone) | Siliceous (granitic) |
| Slope (°) | 10-30 | 15-35 |
| Aspect | North, Northeast | East, Northeast |
| % Surface Rock | 47±30 | 40±28 |
| Mean annual Temperature (°C) | 11.7 | 14.6 |
| Annual Precipitation (mm) | 456 | 764 |

### 3.3. Study design and survey of crops and saplings

In each location, three sites with fifty transects each were established (*Fig.* 4). The transects were selected taking a holm oak tree as a reference and measuring from it 10 meters in the direction to the next reference tree and a width of 2 m (1 meter per side). A minimum distance of 10 meters was established between transects (20 m or more among reference trees). The disposition of the transects of each population was selected forming a pathway, thus long-term records could be done easily.

We recorded crop production of reference trees, natural emergence of new seedlings, and survival of older ones in each transect from 1998 onwards in Montejo de la Sierra (22 years) and from 2001 onwards in Navas de Estena (19 years). Annual surveys were done in October, when acorns have almost finished development and summer abortion and predispersal losses have already acted, as well as when seedling mortality due to summer drought have also ended (Pulido & Díaz, 2005). Crop production was estimated visually on a 0-4 scale (0: no acorns or catkins; 1:<10% of the canopy covered by acorns; 2: 10–50%; 3: 50–90% and 4:>90% (Díaz, et al., 2011; Koenig, et al., 2013). Seedling emergence and survival were assessed by individual identification of every seedling found. New seedlings were mapped and marked permanently with colored wire for long-term monitoring *(Fig.* 5b). Seedlings were distinguished from

resprouts by their color (bright green when alive or dark brown when dead) and by having cotyledons still attached or cotyledon marks if removed by acorn dispersers (Díaz et al. 2011). The state of each seedling (alive, dead -dry-, or absent) has been recorded every year. Holm oak seedlings can dry out and resprout (Pulido & Díaz, 2005), so that marks were not removed from dry seedlings.

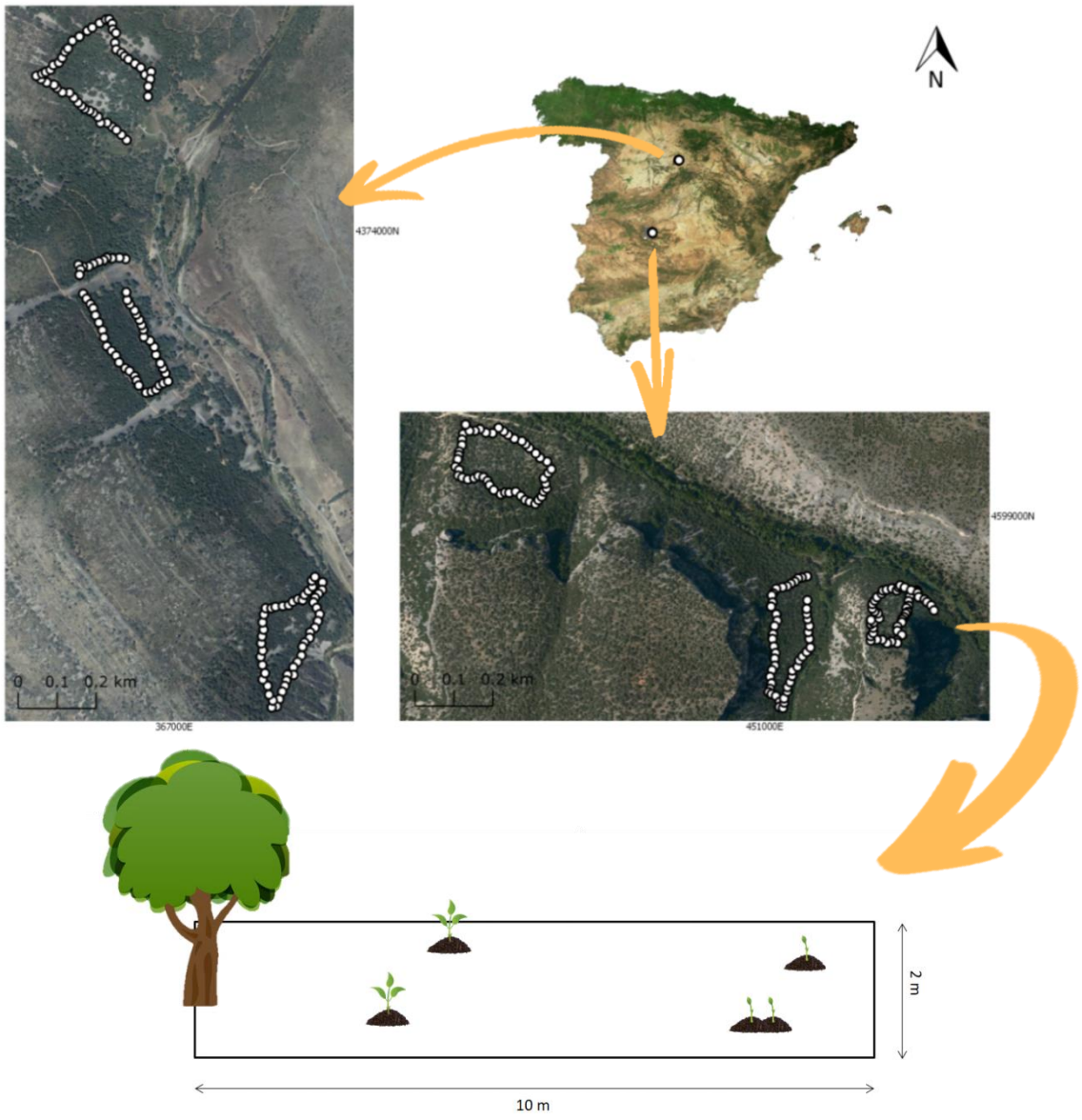


***Figure 4.*** Location of the study

The map on the left corresponds to Navas de Estena (Ciudad Real) and the map on the right to Montejo de la Vega (Segovia). For each location, 3 populations were selected. White circles represent the position of the transects for every population while the draw below represents a transect with its reference tree.

***Figure 5.*** a) Acorns on Holm Oak branches, b) Tagged newly emerged seedlings, note that the acorn remains attached to the plant. c) Delimited transect by a holm oak. Reference trees are marked (tagged) with a metal plate, each surveying period the transects are laid out from the reference tree 10 meters in the direction of the next reference tree.

### 3.3.1. Seedling variables

The emergence of seedlings is measured as the number of newly detected seedlings when alive, per year, and transect; considering than seedlings of a determined cohort will emerge, and thereby will be found, the year after the production of that cohort.

Seedling density was calculated for each year as the number of green seedlings per transect. Dry seedlings were not considered since they are not directly involved in competition or attraction of natural enemies (even if eventually they could resprout). Seedling survival was addressed at the individual level, considering the time interval when they have been alive. Seedlings which

dried out and resprouted were accounted as alive for the whole interval, while the seedling alive at the end of the study (2019) were right-censored. Survival of seedlings of unknown age found in the first census (cotyledon marks may remain from several years; (Díaz, et al., 1997; 2011) was not analyzed, although they were taken into account to compute for seedling density.

### 3.3.2. Crop production and mast years

Crop production of the reference tree of each transect was used to estimate acorn production at the transect scale, assuming a high spatial correlation of acorn crops (results not shown), even higher than temporal synchrony (Koenig et al. 2013). The identification of mast years was done by applying the standard deviate method. The standardized deviation of the annual crop production of each location was calculated from the mean acorn production of the total census. A mast year was considered when the deviate value of a year was, above 1, if not, the year was considered as non-mast.

### 3.3.3. Local vegetation structure

We measured cover of tree and tall shrub canopies when transects were established (1998 or 2001) and in October 2019. Initial estimates were made by eye over 25-m circles per site, located every second reference tree. October-2019 measurements were made for each transect by mapping canopy projections. Both measures were highly correlated (data not shown) after eliminating 8 transects of one of the Navas de Estena sites, that was burned in a large wildfire that occurred in 2012 (seedling data from these transects after burning were removed from analyses). Hence, we estimated canopy covers providing shade to seedlings (and hence protection from early mortality during summer drought; Pulido & Díaz 2005; Smit, et al., 2008) for each transect during the whole study period from measurements made in October 2019.

### 3.4. Climatic variables

Drought is one of the main environmental factors which limits seedling recruitment for Mediterranean *Q. ilex* populations (Pulido & Díaz, 2005; Marañón, 2008; Granda, et al., 2014). The drought index SPEI (*Standardized Precipitation Evapotranspiration Index*) was chosen among other well-supported indexes to estimate drought conditions since it considers the effects of temperature and allows the study at different temporal scales. The SPEI is calculated from the difference of cumulative precipitation for the selected months and potential evapotranspiration (Vicente-Serrano, et al., 2010). High SPEI values indicate wet conditions and low values, drought.

Five initial time-window candidates were tested: SPEI3_P (spring), SPEI3_V (summer), SPEI6_PV (spring and summer), SPEI9_fo (from February to October), and SPEI12_oo (annual, from October to October). The selection of SPEIs was done taking into account Holm oak phenology, as seedling emergence occurs at the end of February, the maximum vulnerability of drought occurs in Spring but mainly in Summer, and that the samplings are censed every October. The SPEI data was obtained from the SPEI global dataset (Appendix 2; Vicente-Serrano, et al., 2010).

### 3.5. Statistical Analysis

#### 3.5.1. First-year survival (Emergence)

Seedling emergence was modeled, at the transect scale, applying generalized mixed models (GLMM) with a negative binomial distribution. The fixed effects of the model were: acorn production, density of conspecific seedlings, tree cover, the SPEI index, and different two-way and three-way interaction of interest (Table 3). Density and climate data were included in the model in t+1 since the emergence of a given cohort occurs the year after the crop production. The final SPEI applied in the models was chosen by correlation of seedling emergence with the SPEI time series (SPEI3_P, SPEI3_V, SPEI9_fo, SPEI12_oo). Finally, the SPEI9_fo, which considers the cumulative drought of the nine months between February and October

The factor "transect" was included as a random factor to account for the repeated nature of the measurements and other unexplained variation. Different random structures were tested, including transect nested into population, and comparisons were made by the likelihood ratio test. The same test was applied for testing the relevance of using random factors.

Initially, Poisson distribution was tested for the construction of the model, however, it was rejected due to high overdispersion. Three alternatives were proposed: Zero-inflated Poisson (ZIB), Negative Binomial (NB), and Zero-inflated Negative Binomial (ZINB). Full models with each one of these distributions were performed and Akaike's information criterion (AIC) was used to determine which model was the most parsimonious.

### 3.5.2. Survival analysis

Survival analyses were conducted using data for the 3344 seedlings of known age, from those, 1094 were found in Navas de Estena and 2250 in Montejo de la Vega. Since the fate of the seedlings alive at the last census is unknown, long-term survival was analyzed by survival curves and hazard models that account for right-censored data.

The Kaplan-Meier method was applied to estimate survival time using log-rank tests (Kaplan & Meier, 1958). Differences in the survivorship curves between mast cohorts and non-mast cohorts were compared following this procedure. Since this method is not appropriate for the study of variables with a high number of levels, quantitative variables, or complex interactions; cox models were used for addressing those variables.

Mixed-effects Cox model, with time-varying covariance, was performed to analyze the effects of conspecific seedling density, tree cover, seedling age, SPEI3_V, status of the cohort (coming from a mast or a non-mast year) and several interactions (Table 3) on survival. The Cox hazard model evaluates the hazard function which indicates the instantaneous risk of mortality at a given time (Cox, 1972). In this study, an extension of the model has been used to allow for time-dependent variables as predictors.

Since the nature of survival data, the SPEI index was chosen by first performing saturated models with each candidate and selecting using the Akaike criterion (AIC), being the SPEI that account for drought in summer (SPEI3_V) the chosen one. Transect was selected as a random factor to account for repeated measurements, following the same procedure as for the emergence model GLMM. Seedling was disregarded as a random factor since not all the seedlings survive for more than one year.

### 3.5.3. Common statistical procedure for GLMM and Mixed-effects Cox model

*Standardization of variables*. To allow direct comparison of effect sizes among predictors, each continuous independent variable was standardized by subtracting the mean of the variable and dividing by its standard deviation.

*Autocorrelation.* Previous to the performance of any model, an analysis of correlation among the explanatory variables was carried out to detect multicollinearity. Since variables did not show a high correlation among them (lower than ±0,15), none of the candidate variables were disregarded.

*Model selection.* Backward elimination was used to select the most parsimonious model. A full model with all the explanatory variables of interest (pure effects and interactions) was first performed, from which alternative models were constructed and compared using their values of AIC (Akaike information criterion). The minimum adequate model was achieved by deleting all non-significant predictors and minimizing the AIC.

*Model validation.* Once the top model was identified, model performance was assessed by inspecting model residuals to confirm that errors were homogeneous and independent for model covariates and location. Model validation, included tests for uniformity of residuals and potential outliers.

Deviance statistics (likelihood ratio test) were applied to assess the significance of random and fixed terms by comparing the final model and the null model one lacking this factor.

*Statistical program and packages.* All analyses were conducted in R (R Core Team, 2019). The package "glmmTMB" (Brooks, et al., 2017) was used to construct GLMMs, the package "survival" and "coxme" (Therneau, 2020a,b) to fit survival curves and survival models, and the "DHARMa" (Hartig, 2020) and "survminer" (Kassambara, et al., 2019) packages for model validation.

## 4. Results

A total of 3571 seedlings were censured over the long-record, from those 1222 were found in Estena (2001-2019) and 2349 in Montejo (1998-2019). After the last census (2019), 89 seedlings remained in Estena and 276 in Montejo (Table 2).

***Table 2.*** *Census data of holm oak emergence in the two areas of study*

| | Montejo de la Vega | Navas de Estena |
|---|---|---|
| Total area sampled ($m^2$) | 3000 | 3000 |
| Recorded years | 1998-2019 | 2001-2019 |
| Total seedlings recorded (number) | 2349 | 1222 |
| Average seedling density (number) | 331.73±303.72 | 174.89±67.66 |
| Average newly emerged seedling (number) | 103.10±281.31 | 42.89±37.71 |
| Tree density ($Ha^{-1}$) | 955.35 ± 511.09 | 485.10 ± 203.12 |
| Average acorn production (%) | 18.47±13.87 | 24.56±10.28 |
| CV acorn production (%) | 75.10 | 41.84 |

*CV: coefficient of variation. Total seedlings recorded including those of known and unknown age.

Mast events occurred in the same years for both locations, in 2003, 2008 and 2015 The 1998 event for Montejo could not be recorded in Estena as monitoring here started later on (*Fig.* 6). "Typical" mast behavior was clearer in Montejo, characterized by a generally lower crop but more variable among years regarding Estena (Appendix 1). The number of newly-emerged seedlings and total seedling density was, in general, larger after years of large crop production,

although in Montejo, some years of high reproductive effort were not followed by subsequent higher emergence, as the mast events of 1998 and 2005 (*Fig* 6b.).

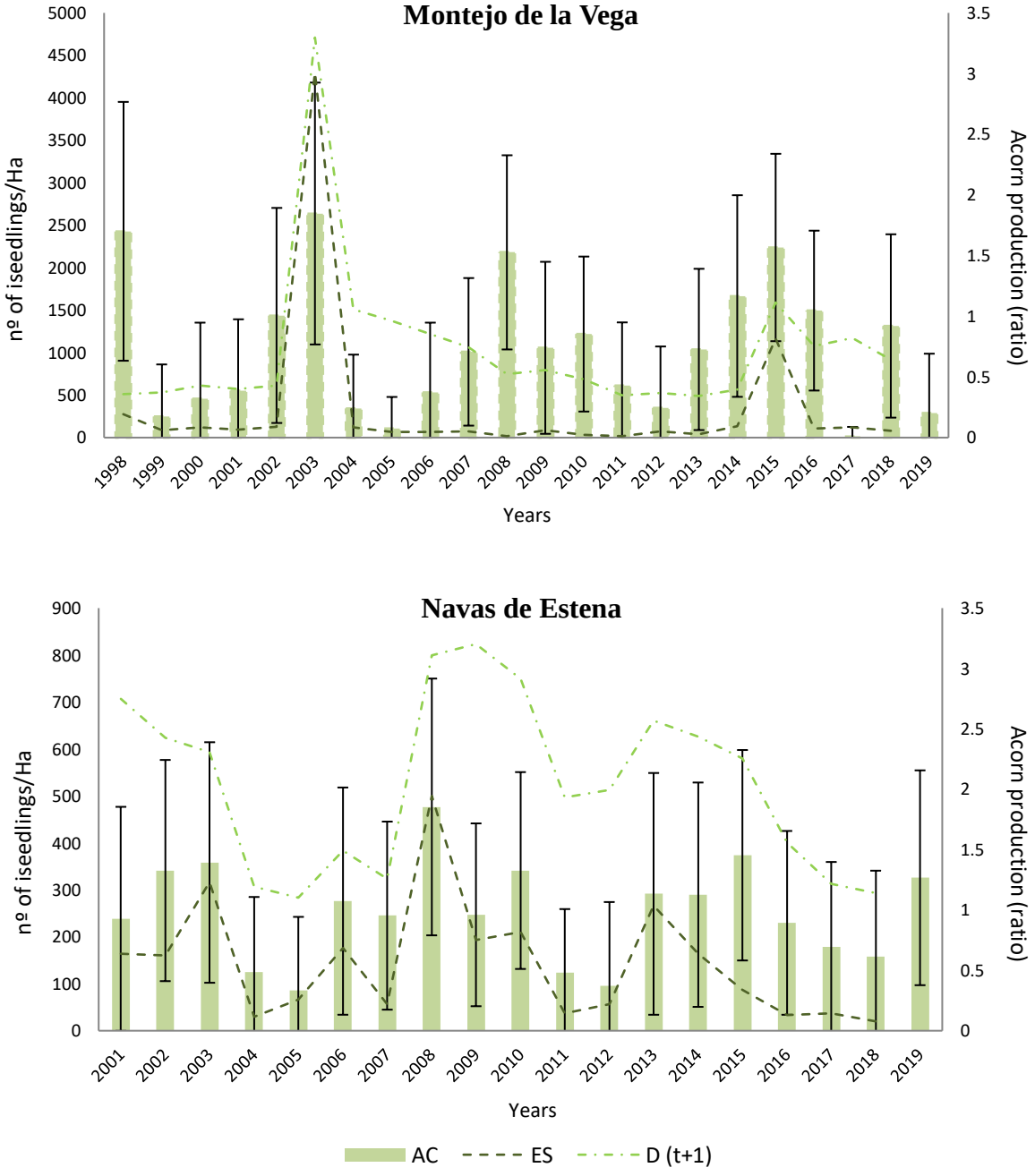


***Figure 6.*** Seedling and acorn production dynamics

AC: acorn production (ratio, 4: total cover, 0: no acorns),

ES: Newly-emerged seedlings; D (t+1): Density of conspecific seedlings in t+1.

Note the difference in the scales of the y-axes (number of seedlings/Ha) between the two locations and the large error bars of acorn production due to large variation among individuals of each population.

Summary of the results obtained in the GLM Negative Binomial model for first-year recruitment and Cox mixed model for long-term survival is shown in Table 3. Whereas the interactions between variables of those models are represented in Figures 7 and 8, for the GLMM and the Cox model, respectively. Results of survival curves are shown in *Figure 9.*

### 4.1. First-year recruitment

Both, acorn production ($z$ = 0.26; $p < 0.001$) and conspecific seedling density ($z$ = 1.89; $p < 0.001$), contributed positively to seedling emergence, although density seems to be the most relevant factor explaining first-year recruitment, especially in Estena ($z$ = -0.86; $p < 0.001$; *Fig. 7b*). Conversely, the effect of crop production was greater in Montejo ($z$ = 0.20; $p < 0.004$ ; *Fig. 7a*). However, when the modulative effect of seedling density over crop production was analyzed, great differences where observed between locations ($z$ = 0.18; $p$ = 0.026; *Fig*. 7h). As was expected, in Montejo, the interactive effect of crop and density had an overall positive effect in recruitment, whereas in Estena the effect of large crops was negative in first-year recruitment at high seedling densities.

In addition, the effects of crop production and seedling density over first-year recruitment were modulated by the local factors tested, which also promoted emergence individually (tree cover: $z$ = 0.14, $p = 0.002$; SPEI: $z$ = 0.23, $p < 0.001$). Thus, the positive effect of acorn production was enlarged by either high tree cover ($z$ = 0.08; $p$ = 0.026; *Fig.* 7d) or high SPEI (wet climate) ($z$ = 0.11; $p$ = 0.003; *Fig*. 7c), although the contribution of the latest was more relevant in Montejo ($z$ = 0.21; $p$ = 0.026; *Fig.* 7i).

Conversely, the positive effect of density on seedling emergence decrease at high tree cover ($z$ = -0.20; $p < 0.001$; *Fig.* 7f). Besides, the modulative effect of seedling density over acorn production also varies with SPEI conditions; with higher SPEI (humid conditions) promoting a stronger negative interaction ($z$ = -0.11; $p < 0.001$; *Fig.* 7g).

## 4.2. Long-term survival

### 4.2.1. Cox model

Masting had shown a negative effect on long-term seedling survival (*HR* = 1.14; $p = 0.014$); however, location was the most important variable affecting hazard ratio, revealing an overall higher risk of mortality in Estena regarding Montejo (*HR* = 0.61; $p < 0.001$). Despite that, in Estena, the increase of mortality associated with masting was attenuated by seedling density (*HR* = 0.84; $p = 0.014$; *Fig*. 8d). In contrast, negative density-dependence effects were predominant in Montejo for both mast and non-mast cohort.

Similarly to first-year recruitment, local factors modulated the effects of masting and seedling density; with low tree cover (*HR* = 0.93, $p = 0.006$, *Fig*. 8c) and SPEI (*HR* = 0.97; $p = 0.005$; *Fig*. 8d) increasing hazard ratio at high seedling densities; and tree cover favoring the survival of seedlings of non-mast cohorts at high seedling densities (*HR* = 1.8; $p = 0.008$; *Fig.* 8e), revealing a more relevant effect of seedling densities for survival when tree cover is lower.

### 4.2.2. Survival curves

Seedling mortality in the first years after emergence declined substantially through the years, but old-seedling was still vulnerable (*Fig.* 9). Kaplan-Meier curves showed a similar pattern for seedling mortality between locations and cohorts than the Cox-model. However, the differences found in the mortality of mast and non-mast cohorts in Montejo seem to be attenuated with the age.

***Table 3.*** *Summary of results obtained from GLMM (Negative Binomial) and COX mixed-model*

| | Estimate | SE | *Z score* | *p value* | | Estimate | HR | SE | *Z score* | *p value* |
|---|---|---|---|---|---|---|---|---|---|---|
| *Emergence model (GLMM with Negative Binomial distribution)* | | | | | *Survival model (COX mixed-model with time-varying covariates)* | | | | | |
| Explanatory variables | | | | | | | | | | |
| Location (Estena vs Montejo) | 0.03 | 0.09 | 0.11 | 0.733 | Location (Estena vs Montejo) | -0.49 | 0.61 | 0.07 | -6.95 | < 0.001 |
| Density (t+1) | 1.89 | 0.10 | 18.34 | < 0.001 | Density | 0.05 | 1.05 | 0.04 | 1.19 | 0.230 |
| Acorn crop | 0.26 | 0.05 | 5.04 | < 0.001 | Cohort (non-mast vs mast) | 0.14 | 1.14 | 0.06 | 2.45 | 0.014 |
| SPEI9_FO (t+1) | 0.23 | 0.04 | 6.13 | < 0.001 | SPEI3_V | 0.001 | 1.00 | 0.07 | 0.07 | 0.980 |
| Tree cover | 0.14 | 0.04 | 3.08 | 0.002 | Tree cover | 0.06 | 1.06 | 0.05 | 0.05 | 0.190 |
| Two-way interactions | | | | | | | | | | |
| Density(t+1): Acorn crop | -0.23 | 0.07 | -3.37 | <0.001 | Density:Cohort | -0.17 | 0.84 | 0.07 | -2.45 | 0.014 |
| Density(t+1):SPEI9_FO(t+1) | -0.07 | 0.04 | -4.64 | 0.09 | Density:SPEI3_V | -0.03 | 0.97 | 0.01 | -2.82 | 0.005 |
| Density(t+1):Tree cover | -0.20 | 0.04 | -4.75 | < 0.001 | Density:Tree cover | -0.07 | 0.93 | 0.03 | -2.77 | 0.006 |
| Acorn crop:SPEI9_FO(t+1) | 0.11 | 0.04 | 3.02 | 0.003 | Cohort:tree cover | -0.08 | 0.92 | 0.05 | -0.12 | 0.270 |
| Acorn crop:Tree cover | 0.08 | 0.03 | 2.48 | 0.026 | | | | | | |
| Location: Acorn crop | 0.20 | 0.07 | 2.84 | 0.004 | | | | | | |
| Location:Density(t+1) | -0.86 | 0.11 | -7.82 | < 0.001 | | | | | | |
| Three-way interactions | | | | | | | | | | |
| Location: Acorn crop:Density(t+1) | 0.18 | 0.07 | 2.48 | 0.026 | Location:Density:Cohort (non-mast) | -0.01 | 0.99 | 0.05 | 0.15 | 0.900 |
| Location:Acorn crop: SPEI9_FO | 0.21 | 0.05 | 4.20 | < 0.001 | Location:Density:Cohort (mast) | 0.14 | 1.15 | 0.06 | 4.44 | 0.025 |
| AC:Density(t+1):SPEI9_FO (t+1) | -0.12 | 0.03 | -3.33 | < 0.001 | Density:Cohort:Tree cover | 0.07 | 1.08 | 0.03 | 2.67 | 0.008 |

SE: standard error. *Z score*: chi-square statistic of the Wald test. HR: Hazard ratio, values above 1 indicate an increase of hazard ratio (mortality risk) and below 1 a decrease hazard.

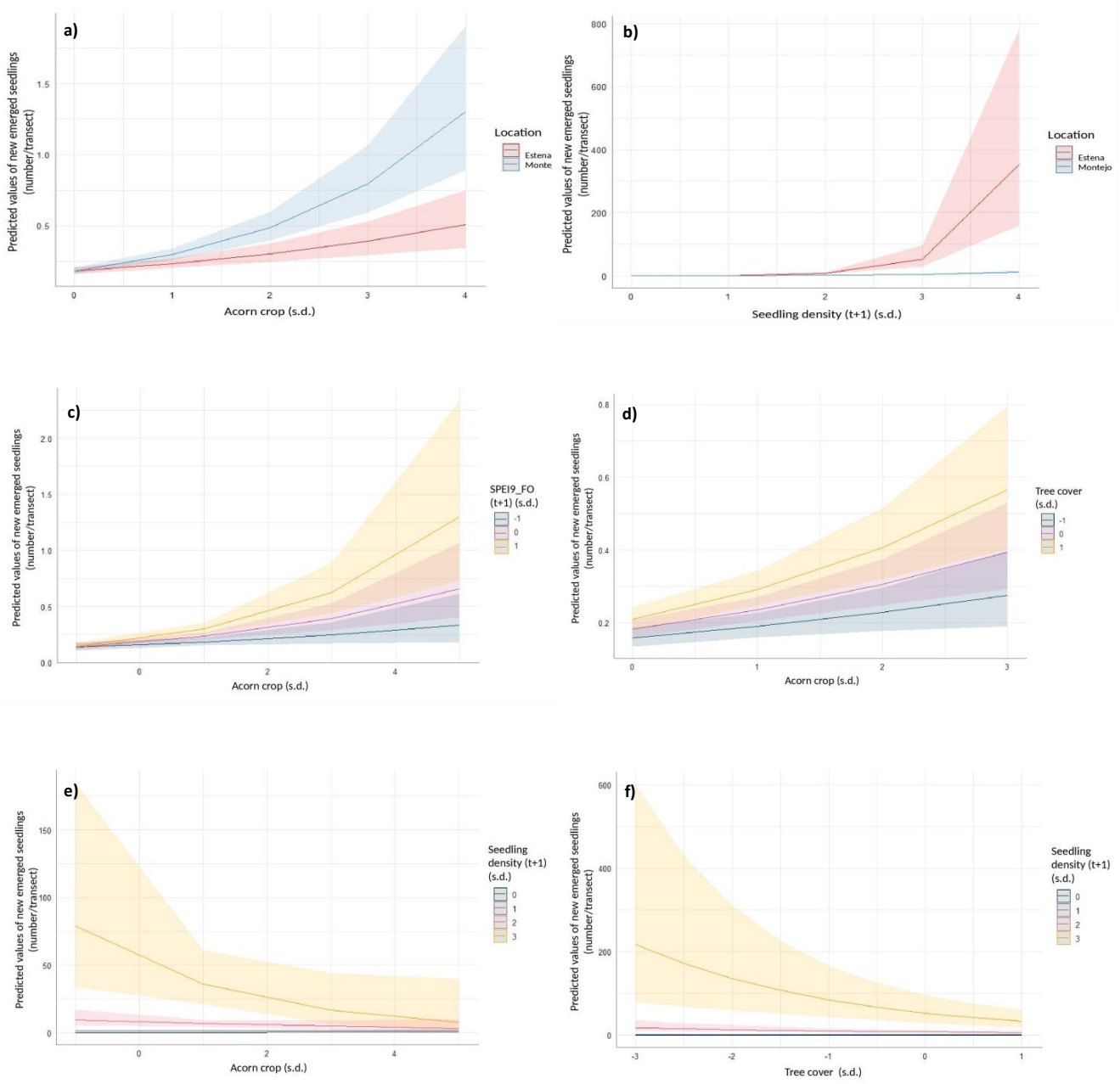


***Figure 7***. Interaction plots of the GLM Negative Binomial Models (emergence)

Plots show the change in the response of one variable in a two or three-way interaction along with their 95% confidence intervals. The response variable (y-axis) is given on the scale of predicted values (marginal effects). Explicative variables were standardized. a) Acorn crop:Location (Montejo de la Vega or Navas de Estena); b) Seedling density (t+1):Location ; c)Acorn crop:SPEI9_FO(from February to October of t+1); d)Acorn crop:Tree cover; e)Acorn crop: Seedling density(t+1); f) Tree cover: Seedling density(t+1).

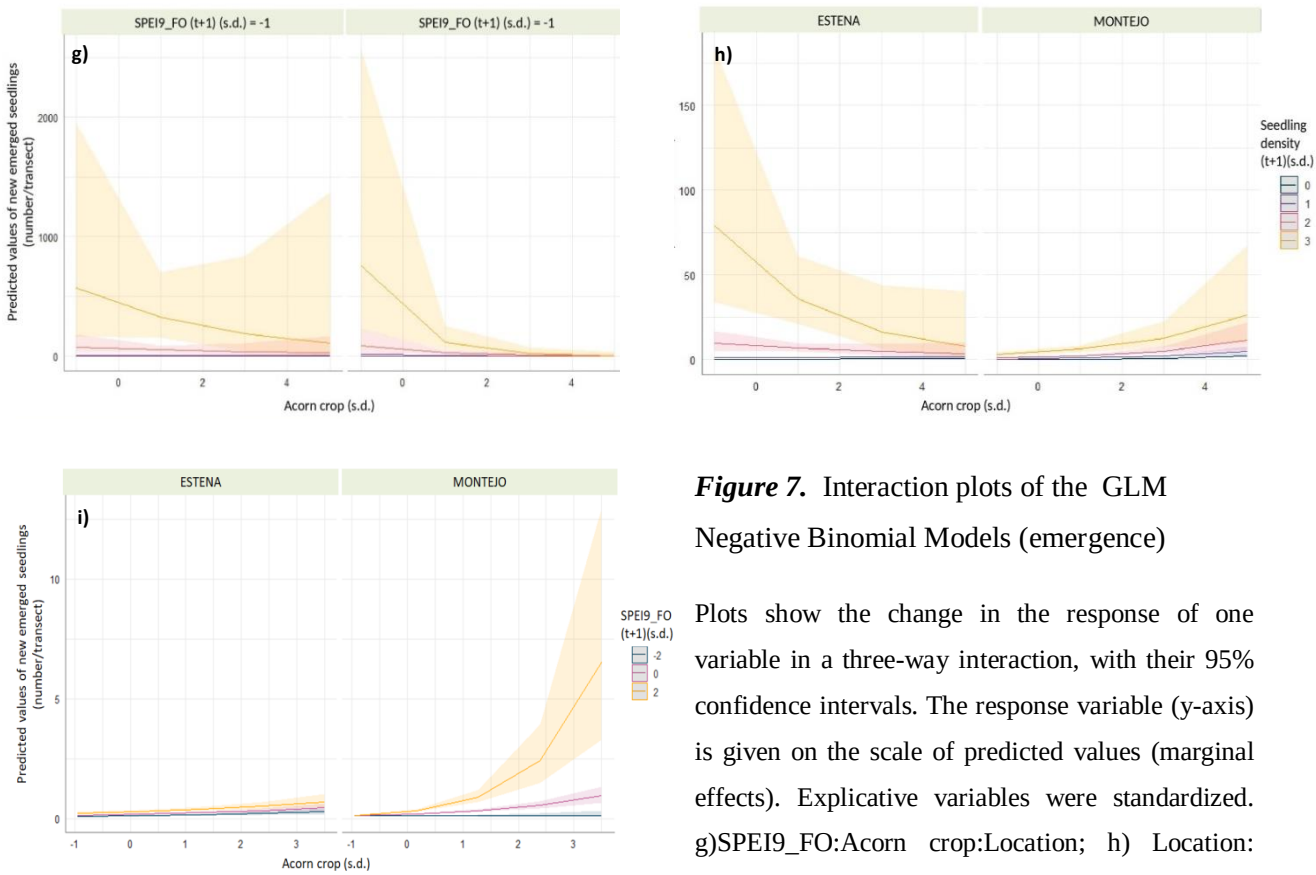


***Figure 7.*** Interaction plots of the GLM Negative Binomial Models (emergence)

Plots show the change in the response of one variable in a three-way interaction, with their 95% confidence intervals. The response variable (y-axis) is given on the scale of predicted values (marginal effects). Explicative variables were standardized. g)SPEI9_FO:Acorn crop:Location; h) Location: Acorn crop: Seedling density (t+1); i) Acorn crop: Seedling density (t+1): SPEI9_FO(t+1).

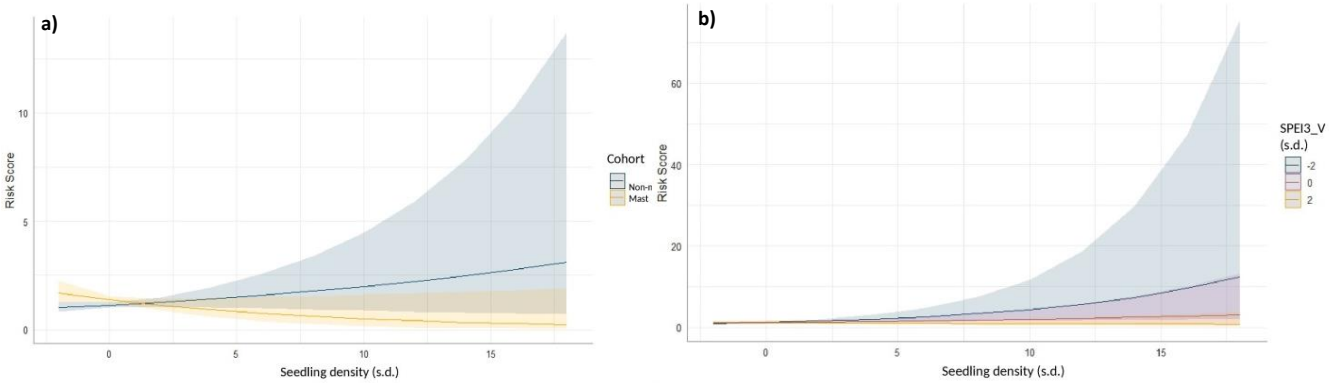


***Figure 8.*** Interaction plots of the Cox hazard model (survival)

Plots show the change in the response of one variable in a two-way interaction along with their 95% confidence intervals. The risk score (y-axis) is given as predicted values. Explicative variables were standardized. a) Seedling density: Cohort (mast or non-mast); b) Seedling density: SPEI3_V (included the 3 months of summer).

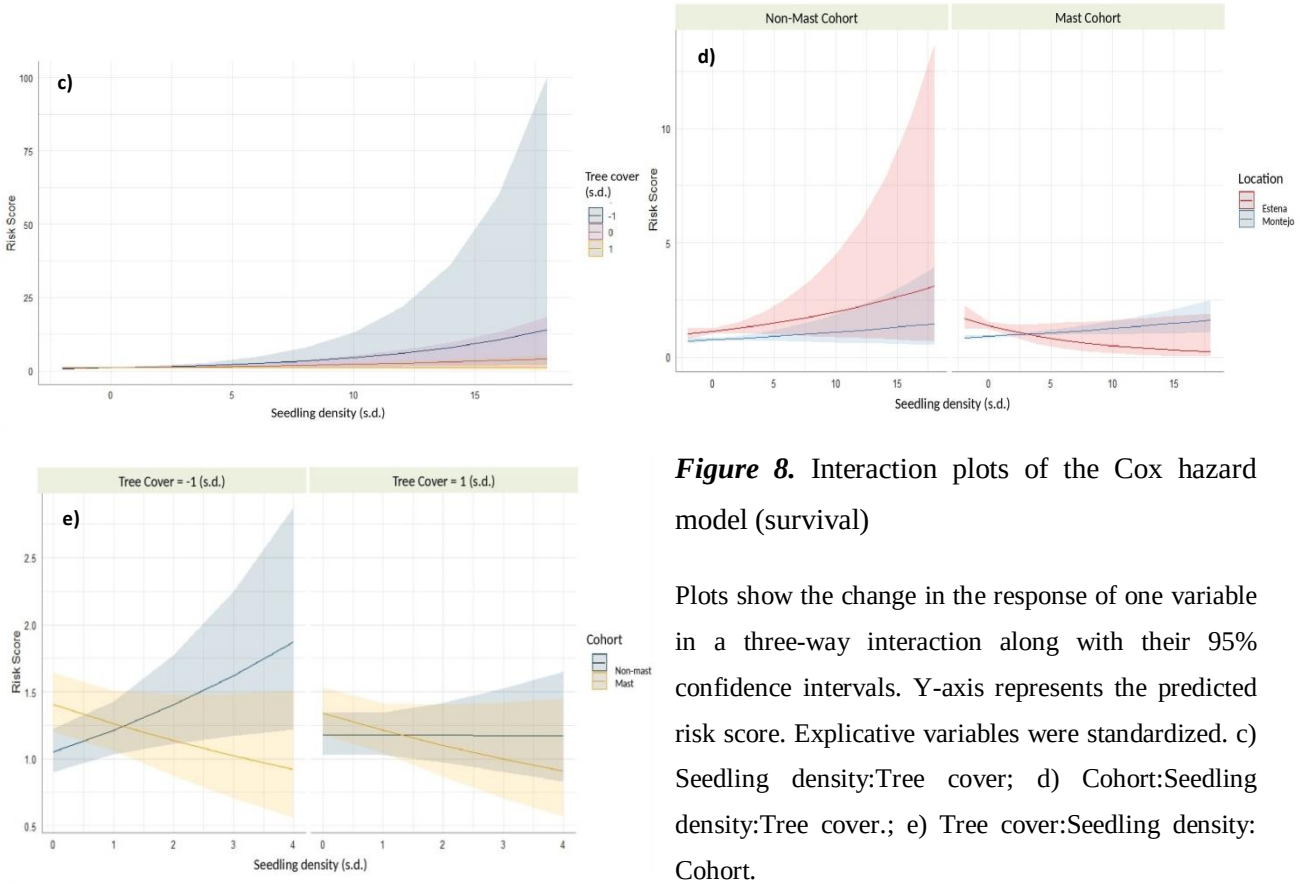


***Figure 8.*** Interaction plots of the Cox hazard model (survival)

Plots show the change in the response of one variable in a three-way interaction along with their 95% confidence intervals. Y-axis represents the predicted risk score. Explicative variables were standardized. c) Seedling density:Tree cover; d) Cohort:Seedling density:Tree cover.; e) Tree cover:Seedling density: Cohort.

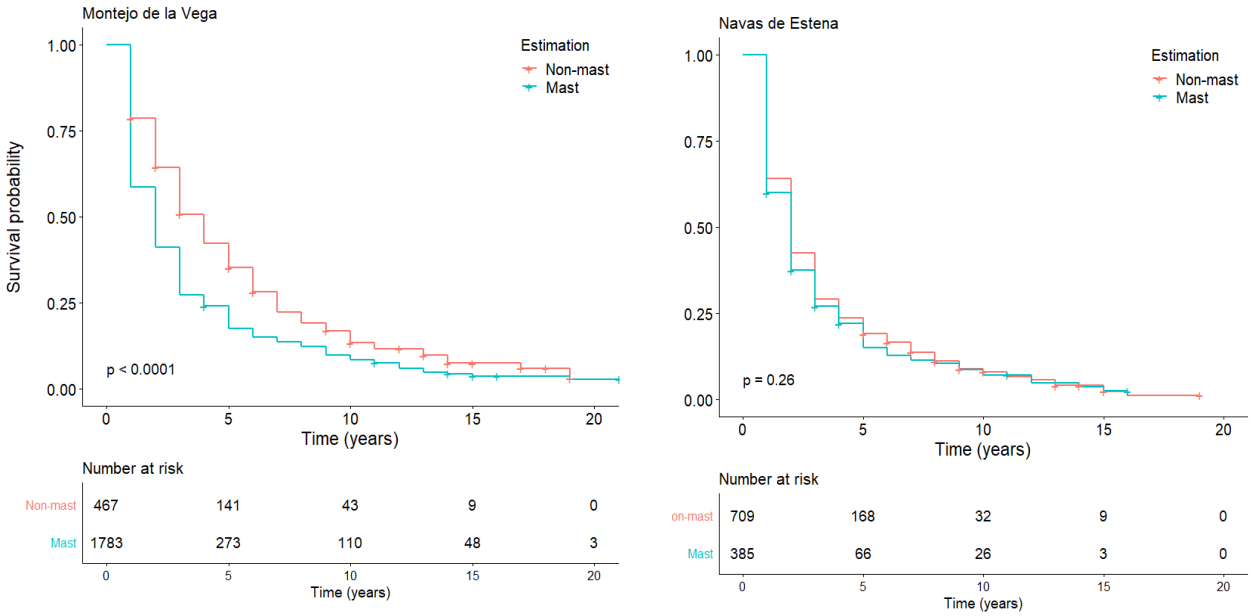


***Figure 9.*** Kaplan Meier Survival curves

*pvalor* calculated by the log-link method.

### 4.3. Seedling populations

As a consequence of the emergence and long-term patterns, the composition of the seedling bank varied from year to year and between locations *(Fig.* 10*)*. The seedling population was more abundant and showed extremer fluctuations in Montejo, where most of the seedlings were originated by mast cohorts. On the contrary, in Estena the number of seedlings was much lower and most of the seedlings came from non-mast cohorts.

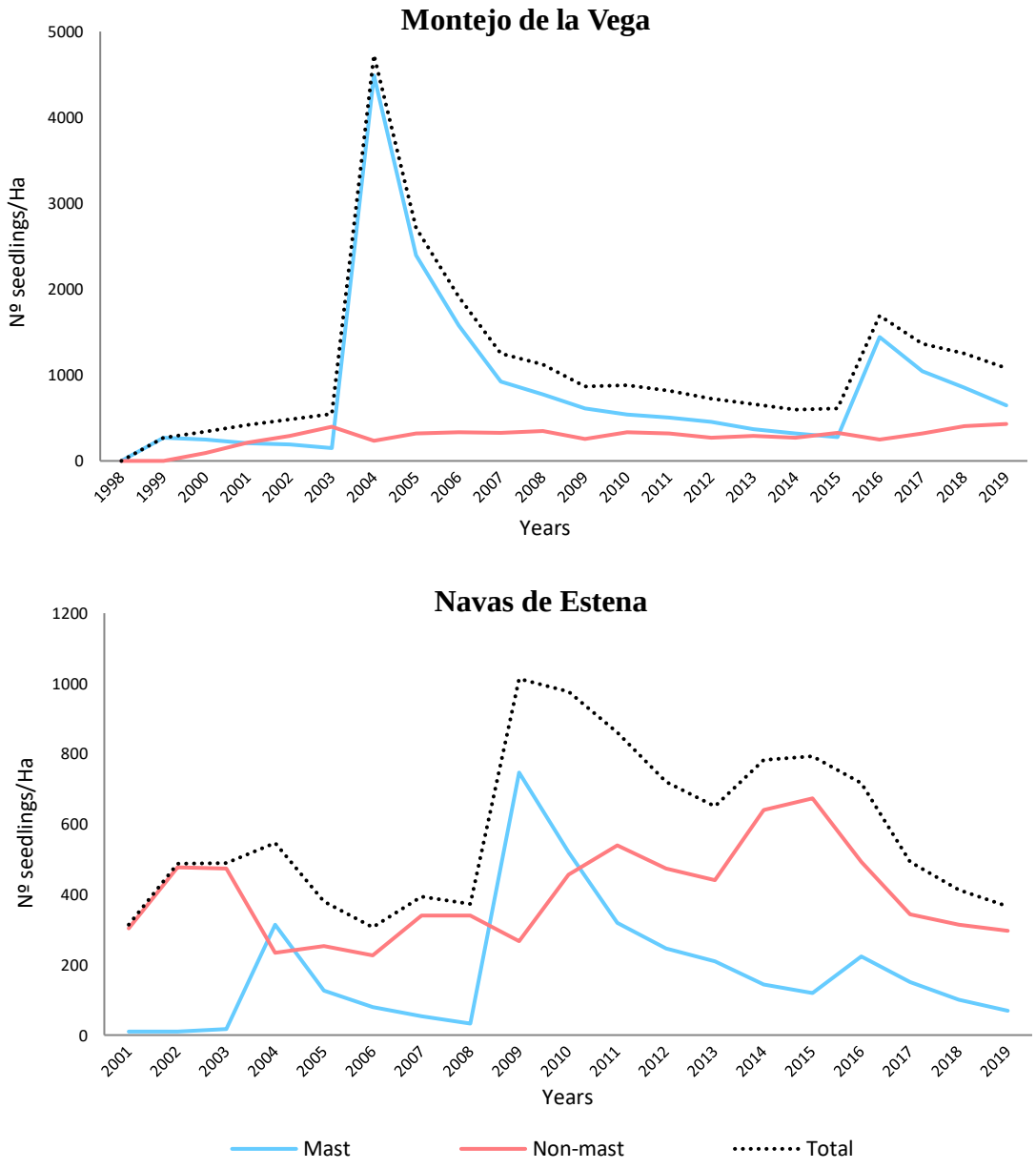


***Figure 10.*** Seedling bank dynamics of the studied locations

# 5. Discussion

## *5.1.* Crop production and masting in *Quercus ilex*

Since the analysis of the proximate causes triggering masting was not one of the goals of this study, and thus not enough data have been recorded to test this idea further (weather cues, explicit acorn production, effects of predator or pollination...) the results obtained of acorn production synchrony and variability will only briefly comment.

Despite the large distance between Montejo de la Vega and Navas de Estena (>300 km), acorn production and the occurrence of mast years at the population level followed a similar interannual pattern. The broad-scale of crop production synchronicity (>100 km) has been documented in several studies, frequently linked to synchrony of weather across large areas (Koening & Knops, 1998; 2013; Schauber, et al., 2002), a phenomenon known as *Moran effect* (Koening & Knops, 2005). As weather drives some key reproductive processes in plants, (e.g. in holm oak, warmer winters seem to be related to earlier budburst, great spring rainfall to pollination failure or summer drought to fruit abortion (García-Mozo, et al., 2007; Espelta, et al., 2008; Pérez-Ramos, et al., 2010), similar weather conditions in both locations during the recorded period could be consistent with the synchronicity of acorn production found. Furthermore, the spatial scale of this synchrony could have been even larger since some similar trends in crop production (e.g. the identification of a mast year in 2003) were found by Carbonero & Fernández-Rebollo (2014) in *dehesas* (holm oak woodlands) of southern Spain (Córdoba).

Estena was characterized by an average higher crop production and lower year-to-year variation, whereas Montejo shows a more typical mast pattern, which seems to be concordant by the hypothesized of Kelly and Sork (2002) of higher population variability at less productive areas, and was also seen by Crone, et al ( 2011) in their comparison among several populations of *Pinus albicaulis* distributed through North America. Masting must be more prevalent in less productive ecosystems where a highly reproductive year constrains the reproduction of successive years as resources linked to reproduction are strongly limited (Lamontagne & Boutin, 2007). Montejo populations are close to the northern edge of Holm oak distribution and are characterized by stronger aridity than Estena populations, which lie closer to the center of

the Holm oak distribution (Blanco, et al., 1997), which could explain the more clear mast behavior in Montejo (*Fig.* 11).

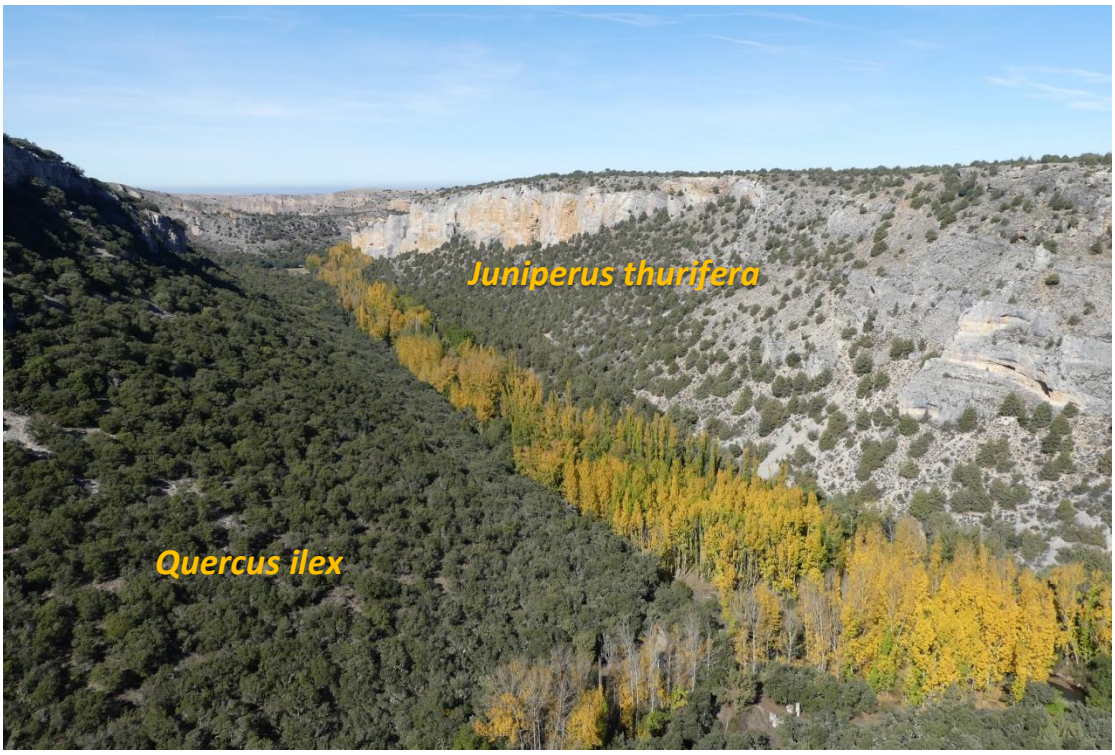


***Figure 11.*** Distribution of *Quercus ilex* in Montejo de la Vega

Holm oak populations in Montejo are replaced by Spanish junipers *Juniperus thurifera* in the drier, south-oriented side of the Riaza river canyon. (Photograph by M. Díaz).

### 5.2. Effects of masting and seedling density on first-year recruitment

Most masting species show at least some reproductive effort between mast events (Sork, et al., 1993; Kelly, 1994), although their relative contribution to total seedling emergence is considered to be much lower than recruits from mast-events (Alvarez-Aquino & Williams-Linera, 2002; Daskalakou, et al., 2019). Despite the large interannual variation in crop production of this study, newly emerged seedlings were found every year. High crop production favored first-year recruitment, especially in Montejo (*Fig.* 6 and 7a); although large recruitment does not necessarily follow mast events (Joshi & Tewari, 2009; Kroiss &

HilleRisLambers, 2015) as several factors are involved in the seed-to-seedling transition (i.e. crop production, seed dispersal, favorable microsites, climatic conditions, predation, etc.; Pulido & Díaz, 2005). Thus, although the larger contribution to seedling emergence during the whole census in Montejo was attributed to the mast events of 2003 and 2015, the also mast event of 2008 was followed by one of the lowest emergences, which may be in part, explained by the drought conditions of this year (Appendix 2). This observation emphasized the importance of the synchronization of the factors involved in successful recruitment.

As well as crop production, conspecific seedling density had a positive effect on first-year recruitment, which could be revealing species-specific habitat preferences that favor recruitment in similar conditions, as opposed to negative density-dependent effects of neighboring seedlings. Several studies have shown than recruitment of several Mediterranean species is limited by the availability of safe or favorable sites (Pulido & Díaz, 2005; Marañón, 2008), where factors constraining seedling recruitment (summer drought, especially are ameliorated; (Pulido & Díaz, 2005; Granda, et al., 2014).

Despite the positive effects of conspecific seedling density could also be attributed to the predator satiation hypothesis (PSH) working on high seedling densities (enhanced by mast events), most studies of PSH in several species including *Quercus ilex*, have shown that its effect is more prevalent at predispersal stages (Espelta, et al., 2008; Bogdziewicz, et al., 2018; Xiao, et al., 2017), since natural enemies are generally host-specific, small and low-mobile, and thereby very susceptible to variability in seed abundance. Contrary, the main potential postdisperal natural enemies in the areas of this study are diet-generalists and highly mobile (jays, wild boars, or rodents) and no-generalized damage by pathogens or predators have been found in the seedlings (although some seedling could have disappeared after the attack), in accordance with other studies of seedling demography in *Quercus ilex* for the Mediterranean area (Pulido & Díaz, 2005).

First-year seedling recruitment seems to be enhanced by both, seedling density and acorn production. Although competition of resources or natural enemies attraction may be still occurring in densely populated sites, the benefits of recruiting in more favorable conditions may outweigh negative density-dependence effects. Nevertheless, the observation found in

Estena of maximum recruitment at higher conspecific seedling densities combined with lower crop productions suggests a possible conflict between seed and seedling stages.

Dense aggregations of holm oak seedlings in favorable sites would probably involve some kind of cover (especially shrubs), to mitigate the drought conditions of summer (Pulido & Díaz, 2005; Granda, et al., 2014). However, vegetation cover may benefit the local activity of seeds predators/dispersers as rodents (Gómez, 2004, Morán-López et al., 2015a, b). Thereby, despite the presence of wild boars, rodents or jays have been found either in Estena and Montejo, their effect could be greater in the first since in Estena first-year recruitment is more limited by favorable sites than to crop production. In this case, safe sites could be favoring negative density-dependence effects at the seed stage but positive effects at the seedling stage. However, further studies on the effects of predators and dispersers on local recruitment on holm oaks in our study sites are thus needed.

***Figure 12.*** Seedlings in a cache

When crop production is abundant, rodents storage the available acorns preferentelly under shrubs forming caches. Frecuently they bury several acorns in the same storage (Gómez, 2004, Morán-López et al., 2015a, b).

### 5.3. Effects of masting and density in seedling survival

Few studies have explicitly compared the long-term survival of seedlings coming from the mast and non-mast cohorts due to the technical difficulties of this approach and the limitation of early recruitment to mast events in most of the species and/or habitats analyzed to date (Alvarez-Aquino & Williams-Linera, 2002). In the present study, the effect of the cohort in survival varied between locations, revealing higher seedling mortality for mast cohorts in Montejo, but

non-significative differences in Estena (*Fig 9*). Extreme declines of seedling density in the subsequent year after a mast event has been largely reported for both temperate (Politi, et al., 2009; Szewczyk & Szwagrzyk, 2010; Daskalakou, et al., 2019) and tropical environments (Alvarez-Aquino & Williams-Linera, 2002), but it is usually assumed that seedling mortality is fairly constant in space and in time after the initial survival bottleneck, giving rise to typical J-shaped age distributions of most tree populations (Condit 1998, Pulido, Díaz y Trucios 2001). However, differences between the mast and non-mast cohorts decline with the age of seedlings (*Fig. 9*) as was also shown in a shorter study (4 years) in *Abies cephalonica* (Politi, et al., 2009).

Larger mortality of mast cohorts seems to be lower at high conspecific seedling densities in Estena, a fact that may indicate that local conditions for early recruitment were also favorable for long-term survival. On the contrary, seedling density seems to increase mortality in Montejo regardless of the cohort, which contrasts with a recent metanalysis showing that negative density-dependence effects were more prevalent in more productive and wetter regions in more productive and wetter regions (Comita et al. 2014).

Results suggest that at least for Estena, areas of high seedling densities are good for both first-year recruitment and seedling survival whereas, in Montejo, the positive effect of density for first-year recruitment does not outweigh the negative effects of high conspecific seedling density (i.e., natural enemies attraction or resource competition (Comita, et al., 2014) in long-term seedling survival. Thereby, recruitment in Montejo is more limited to acorn crop variation which also presents extremer fluctuations; and seedling density could be more related to competitive interactions in future stages than to aggregation in the few good places.

Previous studies analyzing the effect of conspecific seedling density on seedling survival in masting species have found either positive (Calderón-Sanou, et al., 2019), negative (Oshima, et al., 2015) or no effect at all (Martini, et al., 2019). Mixed results were also found in the same studies, with positive or negative effects depending on the mas event (Connell & Green, 2000) or between wet (negative) or dry (positive) season (Lin, et al., 2012). All these results may indicate that the effect of conspecific seedling density determining seedling survival of a masting species is highly dependent on other variables as environmental factors (climatic conditions, vegetation cover, etc…) or intrinsic characteristics of the specie (traits, dominance,

etc), and thus not accounting for local variability may blur patterns of density dependence (Piao, et al., 2013).

### 5.4. Effects of tree cover and climate

Desiccation due to water stress experienced mostly in summer exerts a critical effect on seedling establishment in Mediterranean forests (Pulido & Díaz, 2005; Granda, et al., 2014). Desiccation of seeds may be avoided by either humid conditions or burial by seed dispersers (Gomez, 2004), whereas seedling desiccation risk is reduced by vegetation cover (Pulido & Díaz, 2005; Smit, et al., 2008; 2009) . In both cases, more humid climates and tree cover may help to scape water stress, especially in early stages. Several studies have shown the facilitative effect of some kind of vegetation cover over seedling recruitment and survival in *Q.ilex* (Pulido & Díaz, 2005; Granda, et al., 2014) as a result of amelioration of stressful climatic conditions in Mediterranean ecosystems and also indirectly by inhibition of the herb cover with can be very competitive with young seedlings (Caldeira et al 2014).

Those positive effects of tree cover and humid weather conditions (high SPEI) were also found in this study, mainly in first-year recruitment, playing an important role in modulating the effects of crop production and seedling density. Thus, competition or natural enemies attraction in dense patches is favored when SPEI is high as conditions for first-year recruitment are not as limited to few safe sites (*Fig.* 7g). High tree cover favored long-term survival of seedlings (*Fig*. 8e), and high seedling densities when tree cover or SPEI is low (*Fig*s. 8c and 8b, respectively) favored negative effects of competition or natural enemies attraction on long-term survival.

### 5.5. Seedling fate and evolution of masting

Although several hypotheses have been proposed, a conclusive explanation for masting has not been found yet (Pearse, et al., 2016). In the case of Holm oaks, it seems than masting may have an evolutionary advantage through pollen efficiency (García-Mozo, et al., 2007) and predator satiation hypotheses (Espelta, et al., 2008; Bogdziewicz, et al., 2018), although ultimately crop production is strongly dependent on environmental variability (Pérez-Ramos, et

al., 2010). In any case, masting would provide an evolutionary only advantage if the survivorship of seedlings and their incorporation into the reproductive population is large enough to compensate for later density-dependent mortality (Tachiki & Iwasa, 2010).

During the whole period of study, no-seedling reached the sapling stage. Rather most of them died in the first years after emergence and, although mortality risk decreased with age, most of the seedlings died without never becoming reproductive (Gracia, et al., 2001; Puerta-Piñero, et al., 2012). Seedling mortality was thus a crucial stage for overall recruitment (Granda, et al., 2014) in *Quercus ilex* for the locations here studied.

The initial establishment and survival of *Quercus ilex* seedlings were favored by vegetation cover protecting against drought (Espelta, et al., 1995; Gomez, 2004; Pulido & Díaz, 2005; Marañón, 2008; Smit, et al., 2008); however, seedling growth and subsequent development are limited by light availability (Espelta, et al., 1995; Marañón, 2008; Puerta-Piñero, et al., 2012). Just the formation of a gap in the dense canopy (after a fire, tree fall, or tree cutting) will resolve the conflict between developmental stages, favoring the incorporation to the reproductive population (Puerta-Piñero, et al., 2012). As a consequence, the natural regeneration of this specie will require a certain level of permanent seedling population able to colonize ephemeral habitats available for recruitment (Tachiki & Iwasa, 2010).

In general, the observations of this study show how masting increases the possibilities of first-year recruitment in both locations studied, although it decreases the opportunities for long-term survival in Montejo, but not for Estena.

Masting contributes vastly to the seedling population in Montejo despite the high mortality, whereas non-mast years are still reproductive and contribute to the maintenance of a minimum but constant amount of seedlings in the population. The negative effects of high conspecific seedling density in Montejo on survival may be outweighed if the higher mortality rate is compensated by “frequent” mast events that compensate for the seedling losses.

On the contrary, masting does not seem to be as relevant for seedling populations in Estena. These differences could be in part attributed to the lower differences in crop production between the mast and non-mast years as compared to Montejo, an overall higher acorn production, and a stronger limitation by safe sites for early recruitment.

Thereby, along with this study, the effects of masting and density-dependence have revealed to be highly dependent on local factors for Holm oak. Whereas in Montejo, the less productive and more variable area for crop production but the most prone to seedling competition with pervasive effects over survival, masting could be an advantageous strategy if it occurs frequently enough to maintain levels of seedling density increasing the probabilities of final recruitment. In Estena, at least in the recorded period, masting did not seem to provide an ultimate advantage since early recruitment seems to be more limited by safe sites (shown by the positive effect of seedling density) than to extreme crop production. These observations support the idea than masting is triggered by several factors with are context-dependent.

### 5.6. Limitations of the study

The main purpose of this project was to understand the contribution of masting on plant regeneration through the study of seedling dynamics; a stage frequently underestimated. However, there are important limitations that need to be acknowledged:

Firstly, during the long-term record, some incidents occurred. The most important was a fire in 2012 that affected 5 trees and its transects of the population 1 in Navas de Estena; besides, a tree in this location also died. In both cases records of crop production were used just till the event occurred; although data of the seedlings tagged were still used until its dead. Thus, degrees of freedom for Montejo were fewer than for Estena. Still, since the affection was in a small area we do not think these events affected significatively the results obtained in this study.

Secondly, the "transect" (10m x 2m) has been the main scale of analyses, but the pattern of seedling aggregation and dispersion could vary at finer scales, and thus the effect of seedling density could be underestimated. Nevertheless, with the extensive dataset used (either temporarily and spatially), the results are robust enough to support the main conclusions reached in this study.

Thirdly, since the focus of this study was to understand the density-dependence relations of seedlings, other potentially relevant variables for early recruitment were not included (such as predation, soil, nutrients, or topography), which could be masking patterns of density

dependence or the effects of mast-seeding. However, as previous studies have shown that climate and vegetation cover are the most relevant local factors involved in early recruitment (Pulido & Díaz, 2005), I believe the models including them are accurate enough for the established purposes.

Moreover, the relative contribution of every stage of recruitment could be only completely understood when all of them are analyzed together. Here, as the study was focused on early recruitment and crop production, other relevant stages (i.e. pollination, predispersal, and postdispersal predation or seed dispersión) have not been analyzed. Nevertheless, the analysis of variables more related to fitness than pre-establishment losses allowed to develop evolutionary interpretations of results on stronger grounds than usual.

Finally, despite the long-term approach of this study 20 years is a short time in comparison to the lifespan of trees, as holm oaks, that can reproduce over hundreds of years, and thus final recruitment has not been addressed. Still, some consistent patterns can be highlighted.

## 6. Conclusions

Masting has been proposed to increase seedling recruitment via positive density-dependent effects (Norton & Kelly, 1988). However, the increase of individuals after mast years could lead to stronger interspecific competition among seedlings or the attraction of pathogens or predators (Comita, et al., 2014). In the present study, the effect of masting on early recruitment has been analyzed in two locations of the Iberian Peninsula from 1998-2019 and 2001-2019, showing the following patterns:

In general, masting had a positive effect on first-year recruitment but negative in long term survival, whereas density had an early positive effect (as a result of habitat preferences) but indirect and mixed-effects on long-term survival. However, important differences between locations were found. Density-dependence did not outweigh the initial patterns of masting in Estena (maybe because density is not large enough to show negative interactions but habitat-preferences), whereas in Montejo, seedling density contributes to the greater mortality in the

long term of mast cohorts, probably due to competition and natural enemies attraction in a site where recruitment is less restricted by safe site availability (Comita, et al., 2014).

Climate and tree cover play an important role in modulating the effects of masting and density on early recruitment, but also by themselves, generally ameliorating the pervasive effects of drought and intraspecific competition.

Besides the spatial differences (Montejo vs Estena), the effects of masting and density on early recruitment also show large temporal (first-recruitment vs. survival) variations, as well as the modulative effects of the local factors accounted, tree cover, and climate (drought), reflecting a higher vulnerability at younger stages.

As a consequence of the different factors and interactions involved in early recruitment, the seedling population of Navas the Estena was mostly constituted of seedlings of non-mast cohorts whereas in Montejo the seedlings of mast cohorts were more abundant, which could reflect that frequent mast event could be enough to compensate the population numbers despite the higher mortality in this locations.

As has been shown along with the present study, masting and density-dependence play an important role in the natural regeneration of several species but also in their whole ecosystem through bottom-up effects. In consequence, studies as the present, are fundamental to understand the adaptative ability and resilience of the ecosystem needed for its long-term conservation and sustainable management.

## 7. Agradecimientos

Gracias en primer lugar a Mario Díaz por su esfuerzo como tutor, dedicación, apoyo, y confianza puesta en mí tanto en este TFM como en la realización del TFG, así como a todas las personas que han contribuido a lo largo de los años en el seguimiento de las poblaciones que he podido estudiar. Además, agradezco enormemente a la institución del CSIC por permitirme realizar este TFM bajo el amparo de una beca de investigación JAE, lo cual me ha permitido contar también con las contribuciones y apoyo de todo el Departamento de Biogeografía y Cambio Global.

También me gustaría agradecer al conjunto de profesorado del Máster de Restauración de Ecosistemas por todos los conocimientos y experiencias brindadas a lo largo de este año y por ofrecernos un punto de vista nuevo en cuanto a la gestión y mantenimiento de nuestro legado natural. Finalmente querría agradecer especialmente a todos mis compañeros del máster, por todo lo que me habéis ofrecido tanto en el ámbito académico como personal; estos dos años no hubieran sido ni la mitad de enriquecedores sin vosotros.

## 8. Literature cited

Allen, R. B., Millard, P., Richardson, S. J. 2018. A Resource Centric View of Climate and Mast Seeding. *Progress in Botany* 79:233–268.

Allen R.B., Millard P., Richardson S.J. 2017. A Resource Centric View of Climate and Mast Seeding in Trees. In: Cánovas F., Lüttge U., Matyssek R. (eds) Progress in Botany Vol. 79, Springer, Cham.

Alvarez-Aquino, C., Williams-Linera, G. 2002. Seedling bank dynamics of *Fagus grandifolia* var. *mexicana* before and after a mast year in a Mexican cloud forest. *Journal of Vegetation. Science* 13:179-184.

Archibald, D. W., McAdam, A.G., Boutin, S, Fletcher, Q. E., Humphries, M.M. 2012. Within-Season Synchrony of a Masting Conifer Enhances Seed Escape. *The American Naturalist* 179 (4):536-544.

Beaune, D., Bollache, L., Fruth. B., Hohmann, G., Bretagnolle, F. 2012. Density-dependent effect affecting elephant seed-dispersed tree recruitment (Irvingia gabonensis) in Congo. *Forest. Pachyderm* 52: 97-100.

Blanco Castro, E., Casado González, M.A., Costa Tenorio, M., Escribano Bombín, R., García Antón, M., Génova Fuster, M., Gómez Manzaneque, A., Gómez Manzaneque, F., Moreno Saiz, J.C., Morla Juaristi, C., Regato Pajares, P., Sainz Ollero, H. 1997. Los Bosques Ibéricos: una interpretación geobotánica. Planeta, Barcelona, España.

Bogdziewicz, M., Espelta, J.M., Muñoz, A., Aparicio, J.M., Bonal, R. 2018. Effectiveness of predator satiation in masting oaks is negatively affected by conspecific density. *Oecologia* 186:983-993.

Brooks, M.E., Kristensen, K., van Benthem, K.J., Magnusson, A., Berg, C.W., Nielsen, A., Skaug, H.J., Martin, M. and Bolker, B.M. 2017. glmmTMB Balances Speed and Flexibility Among Packages for Zero-inflated Generalized Linear Mixed Modeling. *The R Journal* 9(2):378-400.

Calderón-Sanou, I., Ríos, L.D., Cascante-Marín, A., Barrantes, G., Fuchs, E.J. 2019. The effect of conspecific density, herbivory, and bamboo on seedling dynamics of a dominant oak in a Neotropical highland forest. *Biotropica* 51(6): 817-825

Carbonero, M. D., Fernández-Rebollo, P. 2014. Dehesas de encinas. Influencia de la meteorología en la producción de bellotas. *Ecosistemas* 23(2):55-63.

Chazdon, R. L., Uriarte, M. 2016. Natural regeneration in the context of large-scale forest and landscape restoration in the tropics. *Biotropica* 48 (6):709-715.

Comita, L.S., Queenborough, S.A., Murphy, S.J., Eck, J.L., Xu, K., Krishnadas, M., Beckman, N., Zhu, Y. 2014. Testing predictions of the Janzen–Connell hypothesis: a meta-analysis of experimental evidence for distance and density-dependent seed and seedling survival. *Journal of Ecology* 102:845–856.

Condit, R., Sukumar, R., Hubbell, S. P., Foster, R. B. 1998. Predicting population trends from size distributions: a direct test in a tropical tree community. *The American Naturalist* 152(4):495-509.

Connell, J. H., Green, P. T. 2000. Seedling dynamics over thirty-two years in a tropical rain forest tree. *Ecology* 81(2):568-584.

Cox, D.R. 1972. Regression Models and Life-Tables. *Journal of the Royal Statistical Society: Series B (Methodological9* 34(2):187-202.

Crone, E. E., McIntire, E. J., Brodie, J. 2011. What defines mast seeding? Spatio-temporal patterns of cone production by whitebark pine. *Journal of Ecology* 99:438–444.

Daskalakou, E. N., Koutsovoulou, K., Ioannidis, K., Koulelis, P.P., Ganatsas, P., Thanos, C.A. 2019. Masting and regeneration dynamics of *Abies cephalonica,* the Greek endemic silver fir. *Seed Science Research* 29(4):227-237.

Díaz, M.. 2014. Distribución del arbolado y persistencia a largo plazo de las dehesas: patrones y procesos. *Ecosistemas* 23: 5-12.

Díaz, M., Alonso, C.L., Beamonte, E., Fernández, M., Smit, C. Desarrollo de un protocolo de seguimiento a largo plazo de los organismos clave para el funcionamiento de los bosques mediterráneos. In: Ramírez, L. y Asensio, B. (eds.) *Proyectos de investigación en parques nacionales: 2007-2010,* pp. 47-75. Organismo Autónomo Parques Nacionales, Madrid.

Díaz, M., Campos, P., Pulido, F. J. 1997. The Spanish dehesas: a diversity of land use and wildlife. In: D. Pain y M. Pienkowski (eds.). *Farming and birds in Europe: The Common Agricultural Policy and its implications for bird conservation.* pp. 178 209. Academic Press, London.

Díaz, M., Pulido, F. J. 2009. 6310. Dehesas perennifolias de *Quercus* spp. In: *Bases ecológicas preliminares para la conservación de los tipos de hábitat de interés comunitario en España,* 69 pp.

Espelta, J. M., Riba, M., Retana, J. 1995. Patterns of seedling recruitment in west Mediterranean mixed holm-oak (*Quercus ilex* L.) forests as influenced by canopy development. *Journal of Vegetation Science* 6(4):465-472.

Espelta, J.M., Cortés, P., Molowny-Horas, R., Sánchez-Humanes, B., Retana, J. 2008. Masting mediated by summer drought reduces acorn predation in Mediterranean oak forests. *Ecology* 89:805-817.

Espelta, J. M., Cortés, P., Molowny-Horas, R., Retana, J. 2009. Acorn crop size and pre-dispersal predation determine inter-specific differences in the recruitment of co-occurring oaks. *Oecologia* 161:559–568.

Frey, B.R., Ashton, M.S., McKenna, J.J., Ellum, D., Finkral, A. 2007. Topographic and temporal patterns in tree seedling establishment, growth, and survival among masting species of southern New England mixed-deciduous forests. *Forest Ecology and Management* 245:54–63.

García-Mozo, H., Gómez-Casero, M. T., Domínguez, E., Galán, C. 2007. Influence of pollen emission and weather-related factors on variations in holm-oak (*Quercus ilex* subsp. *ballota*) acorn production. Envionmental and Experimental *Botany* 61(1):35-40.

Gomez, J. M. 2004. Importance of microhabitat and acorn burial on *Quercus ilex* early recruitment: non-additive effects on multiple demographic processes. *Plant Ecology* 172: 287–297.

Gracia, M., Retana, J., Picó, X. 2001. Seedling bank dynamics in managed holm oak (*Quercus ilex* forests). *Annals of Forest Science* 58 (8): 843-852.

Granda, E., Escudero, A., Valladares, F. 2014. More than just drought: complexity of recruitment patterns in Mediterranean forests. *Oecologia.* 997-1007.

Hacket-Pain, A. J., Lageard, J. G., Thomas, P. A. 2017. Drought and reproductive effort interact to control growth of a temperate broadleaved tree species (*Fagus sylvatica*). T*ree Physiology* 37(6):744-754.

Harper, J. L., 1977. Population Biology of Plants. *Academic Press, London, UK.*

Hartig, F. 2020. DHARMa: Residual Diagnostics for Hierarchical (Multi-Level/Mixed) Regression Models. R package version 0.2.7. https://CRAN.R-project.org/package=DHARMa

Henkel, T. W., Mayor, J. R. 2019. Implications of a long-term mast seeding cycle for climatic entrainment, seedling establishment and persistent monodominance in a Neotropical, ectomycorrhizal canopy tree. *Ecological Research* 472-484.

Henkel, T. W., Mayor, J. R., Woolley, L. P. 2005. Mast fruiting and seedling survival of the ectomycorrhizal, monodominant *Dicymbe corymbosa* (Caesalpiniaceae) in Guyana. *New Phytologist* 167:543-556.

Herrera, C. M. Jordano, P.., López.-Soria, L., Amat, J. A. 1994. Recruitment of a mast-fruiting, bird-dispersed tree: bridging frugivore activity and seedling establishment. *Ecological monographs* 64(3): 315-344.

Hett, J. M. 1971. A Dynamic Analysis of Age in Sugar Maple Seedlings. *Ecology* 52(6):1071-1074.

Janzen, D. H. 1970. Herbivores and the number of tree species in tropical forests. *The American Naturalist* 104:501–528.

Janzen, D.H. 1974. Tropical Blackwater Rivers, Animals, and Mast Fruiting by Dipterocarpaceae. *Biotropia*. 6(2):69-103.

Johnson, D.J., Bourg, N.A., Howe, R., McShea, W.J., Wolf, A., Clay, K. 2014. Conspecific negative density-dependent mortality and the structure of temperate forest. *Ecology* 95(9):2493-2503.

Joshi, B., Tewari, A. 2009. Irregularity in Frequency of Mast Seed Years in Quercus floribunda a Late Successional Species of Central Himalaya. *Russian Journal of Ecology* 40(7):482-485.

E. L. Kaplan, Paul Meier (1958) Nonparametric Estimation from Incomplete Observations. *Journal of the American Statistical Association* 53(282):457-481

Kassambara, A., Kosinski, M., Biecek, P. 2019. survminer: Drawing Survival Curves using 'ggplot2'. R package version 0.4.6. https://CRAN.R-project.org/package=survminer

Keenan, R.J., Reams, Gregory A., Archard, F., de Freitas, J.V., Grainger, A. 2015. Dynamics of global forest area: Results from the FAO Global Forest Resources Assessment 2015. *Forest Ecology and Management* 352(7):9-20.

Kelly, D. 1994. The evolutionary ecology of mast seeding. *Trends in Ecology & Evolution* 9(12):465-470.

Kelly, D., Hart, D. E., Allen, R. B. 2001. Evaluating the Wind Pollination Benefits of Mast Seeding. *Ecology* 82(1):117-126.

Kelly, D., Sork, V. L. 2002. Mast seeding in perennial plants: Why, How, Where? *Annual Review of Ecology, Evolution, and Systematics* 33:427-47.

Kettle, C. J. 2012. Seeding ecological restoration of tropical forests: Priority setting under REDD+. *Biological Conservation* 154: 34-41.

Koening, W.D., Knops, J. M. 1998. Scale of mast-seeding and tree-ring growth. *Nature* 396: 225-226.

Koening, W.D., Knops, J. M. 2005. The Mystery of Masting in Trees: Some trees reproduce synchronously over large areas, with widespread ecological effects, but how and why? *American Scientist* 93(4): 340-347.

Koening, W.D., Knops, J. M. 2013. Large-scale spatial synchrony and cross-synchrony in acorn production by two California oaks. *Ecology* 94(1):83-93.

Kroiss, S. J., HilleRisLambers, J. 2015. Recruitment limitation of long-lived conifers: implications for climate change responses. *Ecology* 96(5):1286-1297.

LaManna, J. A., Walton, M. L., Turner, B. L., Myers, J. A. 2016. Negative density dependence is stronger in resource-rich environments and diversifies communities when stronger for common but not rare species. *Ecology Letters* 19(6):657-667.

LaMontagne, J. M., Boutin, S. 2007. Local-scale synchrony and variability in mast seed production patterns of *Picea Glauca. Journal of Ecology* 95:991-1000.

Li, H.J., Zhang, Z.B. 2007. Effects of mast seeding and rodent abundance on seed predation and dispersal by rodents in *Prunus armeniaca* (Rosaceae). *Forest Ecology and Management* 242:511–517.

Lin, L., Comita, L. S., Zheng, Z., Cao, M., 2012. Seasonal differentiation in density-dependent seedling

survival in a tropical rain forest. *Journal of Ecology* 100(4):905-914.

Marañón, T. Camarero, J., Castro, J., Díaz. M., Espelta, J.M., Hampe, A., Jordano, P., Valladares, F., Verdú, M.. y Zamora, R. 2008. Heterogeneidad ambiental y nicho de regeneración. *In F. Valladares (eds.).Ecología del bosque mediterráneo en un mundo cambiante, segunda edición*, pp. 71-102. Organismo Autónomo Parques Nacionales, Madrid.

Martini, F., Zou, C., Manage Goodale, U. 2019. Intrinsic biotic factors and microsite conditions drive seedling survival in species with masting reproduction. *Ecology and Evolution* 9:14261-14272.

Masaki, T., Abe, S., Naoe, S., Nakajima, A., Nemoto, Y., Yamazaki, K. 2020. Horizontal and elevational patterns of masting across multiple species in a steep montane landscape from the perspective of forest mammal management. *Journal of Forest Research* 25(2): 92-100

Morán-López, T., Alonso, C. L., Díaz, M. 2015. Landscape effects on jay foraging behavior decrease acorn dispersal services in dehesas. *Acta Oecologica 69: 52-64.*

Moreira, X., Abdala-Roberts, L., Linhart, Y. B., Mooney, K. A. 2014. Masting promotes individual- and population-level reproduction by increasing pollination efficiency. *Ecology* 95(4):801-807.

Moreira, X., Pérez-Ramos, I. M., Abdala-Roberts, L., Kailen, A. M. 2016. Functional responses of contrasting seed predator guilds to masting in two Mediterranean oak species. *Oikos* 126(7):1042-1050.

Norton, D. A., Kelly, D. 1988. Mast Seeding Over 33 Years by *Dacrydium cupressinum* Lamb. (rimu) (Podocarpaceae) in New Zealand: The Importance of Economies of Scale. *Functional Ecology* 2(3):399-408.

Oshima, C., Tokumoto, Y., Nakagawa, M. 2015. Biotic and abiotic drivers of dipterocarp seedling survival following mast fruiting in Malaysian Borneo. *Journal of Tropical Ecology* 31:129–137.

Ostfeld, R. S., Keesing, F. 2000. Pulsed resources and community dynamics of consumers in terrestrial ecosystems. *Trends in Ecology & Evolution* 15(6):232-237.

Pearse, I. S., Koening, W. D., Kelly, D. 2016. Mechanisms of mast seeding: resources, weather, cues, and selection. *New Phytologist* 212:546-562.

Pérez-Ramos, I. M., Ourcival, J. M., Limousin, J. M., Rambal, S. 2010. Mast seeding under increasing drought: results from a long-term. *Ecology* 91(10):3057-3068.

Piao, T., Comita, L. S., Jin, G., Kim, J. H. 2013. Density dependence across multiple life stages in a temperate old-growth forest of northeast China. Oecologia 172:207-217.

Politi, P. I., Aruanoutsou, M., Stamou, G. P. 2009. Patterns of *Abies cephalonica* seedling recruitment in Mount Aenos National Park, Cephalonia, Greece. *Forest Ecology and Management* 258:1129-1136.

Puerta-Piñero, C., Pino, J., Gómez, J. M. 2012. Direct and indirect landscape effects on *Quercus ilex* regeneration in heterogeneous environments. *Oecologia* 170:1009-1020.

Pulido, F. J., Díaz, M. 2005. Regeneration of a Mediterranean oak: A whole-cycle aroach. *Ecoscience* 12(1):92-102.

Pulido, F; García, E; Obrador, J, Moreno, J. 2010. Multiple pathways for tree regeneration in anthropogenic savannas: incorporating biotic and abiotic drivers into management schemes. *Journal of Applied Ecology*

47(6):1272-1281.

Pulido, F.J., Díaz, M., Trucios, S.J.H. 2001. Size structure and regeneration of Spanish holm oak Quercus ilex forests and dehesas: effects of agroforestry use on their long-term sustainability. *Forest Ecology and Management* 146(1-3):1-13.

Queenborough, S. A., Burslem, D. F. R. P., Garwood, N. C., Valencia, R. 2007. Neighborhood and community interactions determine the spatial pattern of Tropical tree seedling survival. *Ecology* 88(9): 2248–2258.

Schauber, E.M., Kelly, D., Turchin, P., Simon, C., Lee, W.G., Allen, R.B., Payton, I.J., Wilson, P.R., Cowan, P.E., Brockie, R.E. 2002. Masting by eighteen New Zealand plant species: the role of temperature as a synchronizing cue. *Ecology* 83(5):1214-1225.

Smit, C., den Ouden, J., Díaz, M. 2008. Facilitation of Holm oak recruitment by shrubs in Mediterranean open woodlands. *Journal of Vegetation Science 19:193-200*.

Smit, C., Díaz, M., Jansen, P. 2009. Establishment limitation of holm oak (*Quercus ilex* subsp. *ballota* (Desf.) Samp.) in a Mediterranean savanna - forest ecosystem. *Annals of Forest Sciences 66:511-517*.

Soler, R., Espelta, J.M., Lencinas, M.V., Peri, P.L., Martínez Pastur, G. 2017. Masting has different effects on seed predation by insects and birds in antartic beech forests with no influence of forest management. *Forest Ecology and Management* 400:173-180.

Sork, V. L., Bramble, J., Sexton, O. 1993. Ecology of Mast-Fruiting in Three Species of North American Deciduous Oaks. *Ecology* 74(2):528-541.

Szewczyk, J., Szwagrzyk, J. 2010. Spatial and temporal variability of natural regeneration in a temperate old-growth forest. *Annals of Forest Science* 67: 202.

Tachiki, Y., Iwasa, Y. 2010. Both seedling banks and specialist seed predators promote the evolution of synchronized and intermittent reproduction (masting) in trees. *Journal of Ecology* 98(6): 1398-1408.

Tersago, K., Verhagen. R, Servais, A., Heyman, P., Ducoffre, G., Leirs, H. 2009. Hantavirus disease (nephropathia epidemica) in Belgium: effects of tree seed production and climate. *Epidemiology and Infection* 137:250-256.

Therneau, T.M. (2020). A Package for Survival Analysis in R. R package version 3.1-11. https://CRAN.R-project.org/package=survival

Therneau, T.M. (2020). coxme: Mixed Effects Cox Models. R package version 2.2-16. https://CRAN.R-project.org/package=coxme

Valladares, F., Flores-Rentería, D., Forner, A., Morán-López, T, Díaz, M. 2014. Influencia de la fragmentación y el clima en procesos clave para la regeneración del encinar. *Ecosistemas. 23(2):37-47*.

Vicente-Serrano, S. M., Beguería, S., López-Moreno, J. I. 2010. A Multiscalar Drought Index Sensitive to Global Warming: The Standardized Precipitation Evapotranspiration Index. J*ournal of Climate* 23:1696-1718.

Villar-Salvador, P., Nicolás, J. L., Heredia, N., Uscola, M., 2013. *Quercus ilex* L.In: Pemán J., Navarro-Cerrillo R.M., Nicolás J.L., Prada M.A., Serrada R (eds.). *Producción y Manejo de Semillas y Plantas*

*Forestales*, pp 226-250. Organismo Autónomo Parques Nacionales, Madrid.

Visser, M.D., Jongejans, E., van Breugel, M., Zuidema, P.A., Chen, Y.Y., Kassim, A.R., de Kroon, H. 2011. Strict mast fruiting for a tropical dipterocarp tree: a demographic cost–benefit analysis of delayed reproduction and seed predation. *Journal of Ecology* 99(4):1033-1044.

Waller, D. M., 1979. Models of mast fruiting in trees. *Journal of Theoretical Biology* 80: 223–232.

Xiao, Z., Mi, X., Holyoak, M., Xie, W., Cao, K., Yang, X., Huang, X., Krebs, C.J. 2017. Seed–predator satiation and Janzen–Connell effects vary with spatial scales for seed-feeding insects. *Annals of Botany* 119:109-116.

Xiao, Z., Zhang, Z., Krebs, C. J., 2013. Long-term seed survival and dispersal dynamics in a rodent-dispersed tree: testing the predator satiation hypothesis and the predator dispersal hypothesis. *Journal of Ecology* 101:1256–1264.

Zhu, Y., Comita, L. S., Hubbell, S. P., Ma, K. 2015. Conspecific and phylogenetic density-dependent survival differs across life stages in a tropical forest. *Journal of Ecology* 103(4):957-966.

# APPENDIX 1. DETERMINATION OF MAST YEARS

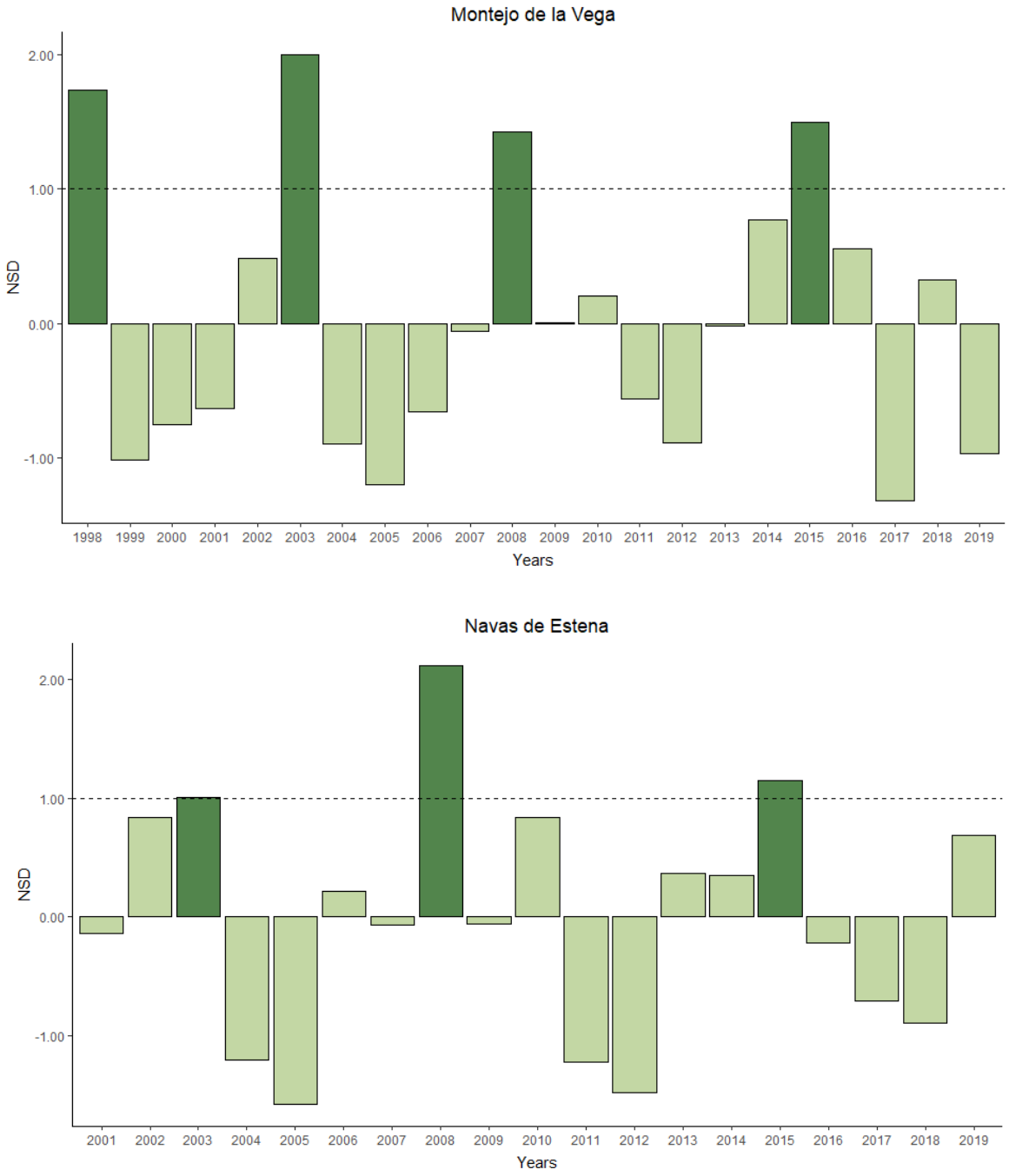


***Figure 1.*** Determination of mast and non-mast years in the two studied locations.

Mast years are calculated in the base of the number of standard deviations (NSD represented in the y-axis) for each year respect to the average acorn crop production in the whole record (per location). When the standard deviation (NSD) reaches the value 1, it was considered a mast-year. For Montejo de la Vega 4 years were identified as mast-years, whereas in Navas de Estena 3 years. *Note that Montejo shows more extreme variations in standard deviations between the mast and non-mast years.

# APPENDIX 2. SPEIs (DROUGHT INDEX)

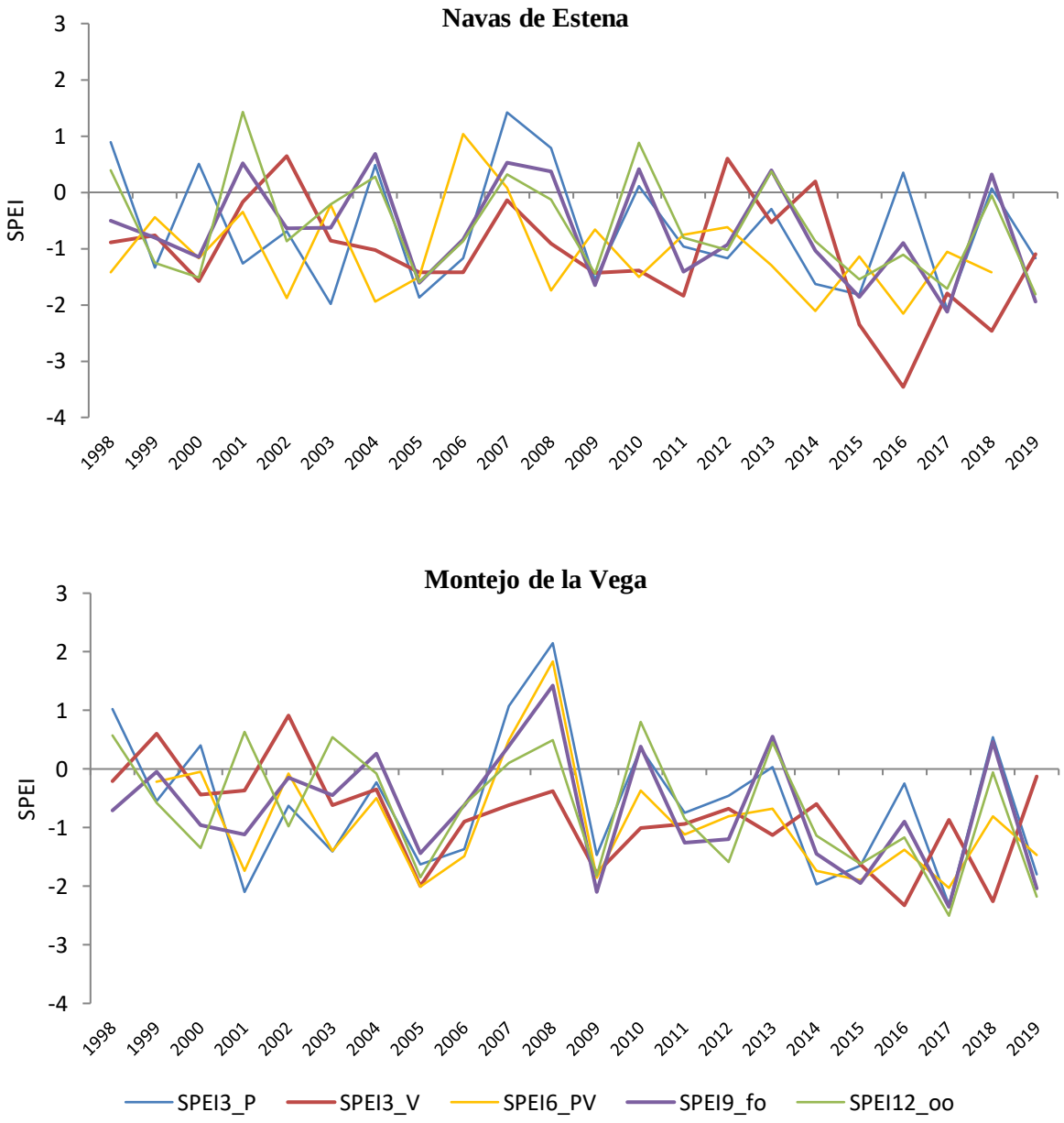


***Figure 1.*** Initial candidates SPEIs tested

SPEI (*Standardized Precipitation Evapotranspiration Index*) is a drought index that considers precipitation and temperature, with the formula: SPEI= P (precipitation) – PET (potential evapotranspiration). Its range oscillates between 4 (wet conditions) and -4 (drought). In the present study, 5 candidates SPEI were studied for its inclusion in the statistical models: SPEI3_P (spring), SPEI3_V (summer), SPEI6_PV (spring and summer), SPEI9_fo (from February to October), and SPEI12_oo (annual, from October to October). Finally, the SPEI9_fo (in t+1) was included in the emergence model (GLMM negative binomial) and the SPEI3_V for the survival model (Cox).