Forest Ecology and Management 598 (2025) 123187

Contents lists available at ScienceDirect

Forest Ecology and Management

journal homepage: www.elsevier.com/locate/foreco

Effect of revegetation method (seedling outplanting *versus* direct seeding) on holm oak root architecture: Implications for restoration success under a global change scenario

Raquel Juan-Ovejero [a,b], Jorge Castro [c,*], Francisco B. Navarro [d], E. Rodríguez-Caballero [e,f], M.P. Reyes-Martín [c], Domingo Alcaraz-Segura [g], M.N. Jiménez [g], Alexandro B. Leverkus [c]

[a] *Department of Ecology and Animal Biology, University of Vigo, Vigo 36310, Spain*
[b] *Centre for Functional Ecology, Associate Laboratory TERRA, Department of Life Sciences, University of Coimbra, Coimbra 3000-456, Portugal*
[c] *Department of Ecology. University of Granada, Granada 18071, Spain*
[d] *Area of Natural and Forest Resources, Institute of Agricultural Research and Training (IFAPA Centro Camino de Purchil), Government of Andalusia, Granada 18004, Spain*
[e] *Agronomy Department, University of Almería, Almería 04120, Spain*
[f] *Research Centre for Scientific Collections from the University of Almería (CECOUAL), Almería 04120, Spain*
[g] *Department of Botany, University of Granada, Granada 18071, Spain*

ARTICLE INFO

Dataset link: Effect of revegetation method (seedling outplanting versus direct seeding) on holm oak root architecture: implications for restoration success under a global change scenario - DATASET

Keywords:
Outplanting
Resprouting capacity
Root branching density
Seeding
Taproot
Terrestrial laser scanning

ABSTRACT

Oaks develop robust taproots that enable them to access water from deeper soil layers and thrive in drought-prone environments. Understanding how different revegetation methods influence oak root development is therefore crucial for improving restoration strategies. In a common garden experiment, we compared the root systems of holm oaks (*Quercus ilex* L.) established through two revegetation methods: seedling outplanting after nursery cultivation and direct acorn seeding. After five growing seasons, we excavated holm oak root systems to a depth of 50–60 cm using a bulldozer and scanned them with terrestrial laser scanning (TLS). Two TLS-derived metrics described the apical dominance gradient of the taproot (index of principal axis dominance and path fraction), while another assessed root ramification (total number of forks per meter). Manual measurements were also taken for taproot diameter, branching root diameters, root branching density, and root:shoot ratio. Moreover, we assessed the resprouting capacity of seeded and planted oaks harvested two years earlier in the same common garden. Multivariate analyses and generalized linear models revealed significant differences between outplanting and direct seeding in root characteristics. Seeded individuals showed greater apical dominance and a higher root:shoot ratio, whereas outplanted ones developed more root ramifications and root branching density, with thicker taproot and branching roots, and a higher ratio of the mean branching root diameter to the taproot diameter. Furthermore, plants from the seeding treatment exhibited a slight but significantly higher resprouting capacity than those from the outplanting treatment. These results indicate that direct seeding promotes a more natural root structure, with stronger taproots and better resprouting capacity than outplanting. Our findings suggest that direct seeding may provide a more nature-based solution than outplanting for forest restoration by emulating natural regeneration. In the mid-term, the root development shown by direct acorn seeding may enhance the ability of holm oaks to withstand disturbances such as droughts in the Mediterranean Region under global change.

1. Introduction

Restoring forests has never been more critical than now, given the intensifying impacts of climate change, biodiversity loss, and the increasing degradation of ecosystems (Lewis et al., 2019; Hua et al., 2022; Mansourian et al., 2024, 2025). Approximately 2 billion hectares of land are suitable for forest restoration (Laestadius et al., 2011; Cernansky, 2018), offering a key opportunity for large-scale initiatives

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* Corresponding author.
E-mail address: jorge@ugr.es (J. Castro).

https://doi.org/10.1016/j.foreco.2025.123187
Received 20 May 2025; Received in revised form 14 September 2025; Accepted 16 September 2025

(Bastin et al., 2019). However, high costs and misguided technical decisions present challenges that can compromise the success of these efforts (Austin et al., 2020; Castro et al., 2021; Cook-Patton et al., 2021). Revegetation plays a crucial role in stabilizing soils (e.g., Scotton and Andreatta, 2021), regulating the water cycle (e.g., Trabucco et al., 2008; Keller and Fox, 2019), and enhancing carbon sequestration (e.g., Walker et al., 2022; Ménard et al., 2023), among other benefits. In areas where natural regeneration is blocked or too slow, active interventions such as outplanting and direct seeding become essential (Ceccon et al., 2016; Grossnickle and Ivetić, 2017). However, for such actions to be effective, we need to carefully select appropriate revegetation methods to ensure plant survival and growth (Lázaro-González et al., 2023 and references therein; Preece et al., 2023).

After revegetation, proper root development underlies the plant's capacity for water and nutrient uptake (Freschet et al., 2021). For species like oaks (*Quercus* spp.), whose extensive root systems allow them to thrive across diverse environments, root growth is a requisite for long-term restoration success (Gil-Pelegrín et al., 2017). Oaks are well-known for their ability to access water and nutrients from deep soil layers, an adaptation for surviving in nutrient-poor and drought-prone environments (Osonubi and Davies, 1981; Padilla and Pugnaire, 2007). Their deep taproots enhance resilience to abiotic stressors, enabling stable growth even under challenging climatic conditions, including prolonged droughts and high temperatures (Barbeta and Peñuelas, 2017). Equally important, robust root systems sustain a strong resprouting ability, and in Mediterranean and other disturbance-prone ecosystems, resprouting is one of the most reliable persistence strategies among angiosperms, allowing trees to recover after damage (Pausas and Keeley, 2014). In oaks, this capacity is crucial for survival and demographic stability in the face of recurrent disturbances such as wildfires, herbivory by ungulates and other large animals, pest outbreaks, multiple forms of management, and extreme climatic events (Sakai et al., 1997; Pyttel et al., 2013). Resprouting enables oaks to maintain high functional levels even under severe stress by rapidly regenerating shoots from protected buds and well-resourced root systems (Espelta et al., 2003). In the context of forest restoration, deep rooting and vigorous resprouting are thus decisive for long-term success. Both, however, can be strongly influenced by the revegetation method, i.e., whether trees are established after nursery cultivation or through the direct seeding of acorns (Castro et al., 2015; Löf et al., 2019).

Nursery-grown oak seedlings are typically cultivated for one year or less in seedbeds or small containers before being outplanted in the field (Pemán et al., 2006; Wilson et al., 2007; Chirino et al., 2008). This method tends to result in larger initial plant size compared to direct seeding and it avoids the risk of demographic bottlenecks during seed predation and germination and seedling early growth (Dey et al., 2008). However, the confined conditions in nursery containers restrict root development, leading to issues such as root deformities, which can limit the formation of a deep taproot and impair water uptake (Tsakaldimi et al., 2009; Mariotti et al., 2015; Grossnickle and Ivetić, 2022). These deformities can reduce access to soil resources, thereby increasing water stress and compromising long-term plant performance in the field (Zadworny et al., 2014, 2019). In contrast, direct seeding of acorns may allow for more natural root development, fostering the growth of deeper taproots that are better suited for accessing water and nutrients (Pemán et al., 2006; Tsakaldimi et al., 2009; Castro et al., 2015; Löf et al., 2019). Direct seeding offers additional advantages, such as lower cost, lower difficulty, greater flexibility in terms of timing and planning, and reduced risk of introducing pathogens through nursery stocks (Fernández-Habas et al., 2019; Lázaro-González et al., 2023). Moreover, for *Quercus* and more generally for large-seeded species, direct seeding has often resulted in higher establishment success than outplanting (Löf et al., 2019).

Despite the well-documented differences in early development and the relevance of root development for a better accessibility to soil resources, the relationship between revegetation method, root system architecture, and resprouting capacity has not yet been evaluated in tree saplings under experimental field conditions. Previous studies addressing this question have largely focused on seedlings or very young plants, whereas little is known about how these processes occur in fully developed trees several years after establishment. In particular, research on the response of oaks to revegetation methods has mostly concentrated on short-term responses (i.e., 1 year old trees or even less), and particularly on survival and aboveground parameters such as growth and biomass accumulation (Lázaro-González et al., 2003). Root traits have also been examined, but predominantly at very early developmental stages (e.g., Pemán et al., 2006; Tsakaldimi et al., 2009). To address this gap, we focused on the holm oak (*Quercus ilex* L.), the most widely distributed tree in the Mediterranean region and highly adapted to summer drought, nutrient-poor soils, and high temperatures (Navarro-Cerrillo et al., 2018; Martín-Sánchez et al., 2022; Juan-Ovejero et al., 2024), which makes it a key species for Mediterranean forest restoration (Leverkus et al., 2015a,b). We analyzed the architecture of the taproot system and its main branching roots (i.e., those coarse roots emerging directly from the taproot rather than fine roots) as well as resprouting capacity in *Q. ilex* five years after establishment under contrasting revegetation methods, thereby offering new insights into their implications for forest restoration. Moreover, we combined advanced terrestrial laser scanning (TLS) and manually-measured variables to provide a detailed analysis of the root architecture of *Q. ilex* individuals from experimental outplanting and seeding treatments. We hypothesized (i) that holm oaks established through seeding would exhibit a stronger dominance of the taproot compared to planted individuals. We also hypothesized (ii) that seeding would result in an enhanced resprouting capacity compared to planting as a result of an enhanced ability to access resources. We seek to improve our understanding of oak root morphology and help optimize revegetation strategies in the Mediterranean Basin by balancing the trade-offs between seedling outplanting and direct acorn seeding.

2. Materials and methods

2.1. Study location and experimental design

A common garden experiment was established in December 2017 at the IFAPA-Camino de Purchil Research Station (37º10'20'' N, −3º38'39'' W; 625 m a.s.l.; Granada, southern Spain) to investigate how revegetation method (seedling outplanting *versus* direct acorn seeding) influences plant performance and the success of reforestation with the holm oak. The site is characterized by a Mediterranean climate, with hot, dry summers and rainfall mainly in autumn and spring. The mean annual precipitation is 389 mm, and the mean annual temperature is 15.3 °C. January is the coldest month, with an average temperature of 6.2 °C, whereas July is the warmest, with an average temperature of 26.0 ºC (data extracted from the IFAPA meteorological station; period 2006–2021). The soil is calcaric fluvisol, with an average of 40 % sand, 18 % clay, 17 % coarse loam, and 25 % fine loam in the first 20 cm of the profile. More detailed information of the characteristics of the soil profile up to 1 m depth is shown in Table S1.

The experimental setup of the common garden site included five adjacent blocks measuring 56 × 28 m, each with 384 planting points arranged in a regular 2 m grid (Fig. 1). Within each block, 192 points were randomly assigned to nursery-grown seedlings (outplanting treatment, hereafter), and the remaining 192 were allocated to direct seeding of an acorn in the field (direct seeding treatment, hereafter). In December 2017, the experiment was established by transplanting the nursery-grown plants (cultivated from February to December 2017; outplanting treatment) and by directly sowing acorns in the field (direct seeding treatment, hereafter). Acorns for the outplanting tratment were collected in autumn 2016 from four provenance populations located in distant regions of the Iberian Peninsula with contrasting climatic conditions, elevations, and bedrock types (see Juan-Ovejero et al. 2024 for

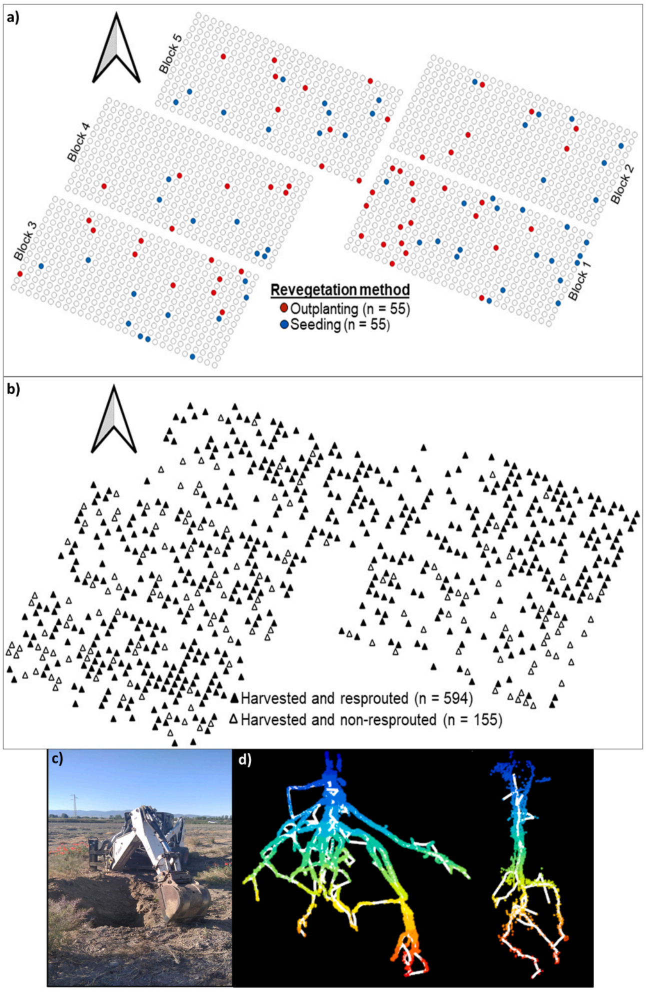

Fig. 1. a) Layout of the common garden experiment with the 110 excavated root systems (outplanting in red and seeding in blue; 104 were used for analyses), b) Harvested and resprouted (black triangles; n = 594 excluding excavated individuals) and non-resprouted plants (white triangles; n = 155), c) Root extraction with heavy machinery, and d) Terrestrial laser scanning of the root systems: left, a root from the outplanting treatment; right, a root from the seeding treatment.

more details). For each population, a similar number of acorns from 3 maternal plants were harvested and later cultivated until Autumn 2017 in a commercial nursery (Viveros Ponce Lajara; 37º46'18.18'' N, −2º32'34.19'' W; 864 m a.s.l.; Galera, Granada, southern Spain) under standard nursery conditions in 300-ml containers filled with a Pindstrup substrate (peat:vermiculite 1:1 v:v), coconut coir dust, slow-release fertilizer (NPK 15:9:11) and conventional irrigation. Acorns for the seeding treatment were collected in Autumn 2017 in the same populations from another 3 maternal plants per population, with a similar number per maternal plant. In December 2017, the experiment was established by transplanting the nursery-grown plants (therefore cultivated in the nursery for one-growing season; outplanting treatment) and by directly sowing acorns in the field (seeding treatment). Outplanted plants had a leader shoot length of 24.10 ± 0.28 cm at the moment of the experimental set up. Weeds were periodically removed from the entire experimental area using manual methods and a cultivator. The area is fenced and therefore free from large mammals such as wild boar or ungulates. Herbivory by ungulates is therefore absent. More detailed information about the experimental set up of the common garden can be found in Juan-Ovejero et al. (2024).

2.2. Root excavation and resprouting assessment

In autumn 2021 (October and November), the aboveground portion of 859 holm oaks distributed within the common garden was harvested, including 617 plants from the outplanting treatment (that were 5-year old at the moment of harvest, 4 in the field plus 1 in the nursery) and 242 from the seeding treatment (that were 4-year old). Before harvesting, the leader shoot length of all plants was measured, and the shoot biomass was also determined after oven-drying at 60 ºC to constant weight. Leader shoot length and shoot biomass were greater in outplanted holm oaks (96.62 ± 1.95 cm and 638.81 ± 35.88 g) compared to seeded individuals (59.75 ± 2.61 cm and 147.02 ± 16.15 g). Statistical analysis revealed significant differences between revegetation methods in both leader shoot length ($F = 110$, p-value < 0.001) and shoot biomass ($F = 71.41$, p-value < 0.001). These data were used to analyze resprouting capacity (see below).

Later, in spring 2022 (May), 110 resprouted individuals from the previously harvested ones were randomly selected for excavation (55 from the outplanting treatment and 55 from the direct seeding treatment; Fig. 1a). A bulldozer was used to extract the roots to a depth of 50–60 cm (Fig. 1c). Root systems were collected randomly across blocks, using a similar number of plants per treatment and block, so potential edaphic effects can be reasonably excluded from influencing our results. The root systems within this depth were carefully removed, transported to a warehouse, and thoroughly cleaned using a brush. The root excavation was carried out six months after the harvest of the aboveground part of the plant, and the time elapsed between aboveground harvest and root excavation corresponded mostly to the winter, a dormant phase for holm oak in the region. Moreover, all the root systems excavated corresponded to individuals that were initiating resprouting, and thus they were live individuals (resprouting capacity was in any case very high, see Results). Altogether, this supports that aboveground cutting six months before root extraction would not affect the root architectural parameters analyzed in this work. Of the 110 excavated root systems, 104 were retained for analysis based on their minimal root damage (52 from outplanting treatment and 52 from seeding treatment).

In spring 2023 (two years after cutting), the resprouting capacity was assessed on the non-excavated plants by identifying which of the individuals harvested in 2021 had successfully produced new shoots ($n = 749$, excluding the 110 individuals whose root systems were extracted; Fig. 1b). The leader shoot length of all plants was measured again at this stage.

2.3. Root architectural metrics and terrestrial laser scanning (TLS)

Once washed and cleaned of all soil material, the 104 excavated roots were brought to laboratory for data acquisition. Both manual and scanned data were collected. First, we manually measured taproot length at the cut made by the bulldozer, the number of coarse branching roots emerging from the taproot (ranging from 0 to over 15), and the diameters of both the taproot and branching roots, with all measurements done at 1 mm precision. Taproot diameter was determined using two perpendicular measurements at the base of the collar, while branching root diameter was measured similarly at the point of insertion with the taproot. Root branching density was calculated as the number of branching roots per meter, and the ratio of the mean branching root diameter to the taproot diameter was also determined. Additionally, the 104 root systems were weighed, and the root:shoot ratio was calculated as the ratio of the total weight of the roots to the weight of the shoot biomass harvested in autumn 2021. We assume that significant changes in the weight of the coarse roots extracted for our study did not happen during the period elapsed from aboveground harvest to root excavation. In any case, any potential effect may affect similarly all the plants, and the data of root:shoot ratio were used for comparisons among revegetation treatments.

Later, roots were scanned using terrestrial laser scanning (TLS) technology (Fig. 1d). A Leica ScanStation 2 terrestrial laser scanner was employed to capture 3D point clouds of the root systems (0.005 m distance between points) from multiple angles to minimize occlusions. This system operates in the green region of the spectrum (532 nm) and has a maximum range of 300 m (at 90 % albedo), laser spot size of 6 mm, beam divergence of 0.15 µrad, and accuracy and precision of 4 and 2 mm, respectively. The scanning process took place over four sessions across three days. The different point clouds of each session were merged using four targets common to both scans with the Cyclone 7.1 software (Leica Geosystems).

Each root system was manually extracted from the merged TLS point cloud and used to develop a quantitative structure model (QSM). First, the point cloud was divided into 2 cm-thick rectangular layers. These layers were then segmented into distinct clusters based on point distances, with points that exceeded a specified threshold (i.e., defined as the average distance of the points of the previous cluster to the center of their corresponding cluster) considered separate objects. Cluster centers were subsequently used as nodes to iteratively construct the root system skeleton, and the radius of each segment was determined. Finally, root topology was established by identifying 'mother' and 'daughter' segments, the paths (i.e., defined as a continuous sequence of segments extending from the tip of a branching root to the taproot), ramifications (i.e., forks), and branching root angles.

Obtained QSMs were used for the calculation of different TLS-derived root architectural metrics related with the dominance of the principal root axis and the ramifications of roots following the methodology described by Martin-Ducup et al. (2020).

We first calculated the number of paths of each root system. Because the finest root tips were likely lost during excavation with the bulldozer (the smallest radio of the different segments conforming each root system was 0.003 m), we quantified root branching architecture following the approach of Martin-Ducup et al. (2020), recording the number of branching paths as a continuous variable from the first visible segment (even if cut) down to the taproot. Afterwards, the length of the different paths found at each root system was calculated, and used to estimate the path fraction (Pf) as indicator of the apical dominance gradient of the taproot (Smith et al., 2014):

$$Pf = \frac{\overline{L_{\updownarrow}}}{L_{\updownarrow}} \tag{1}$$

where$\overline{L_{\updownarrow}}$ is the mean path length and $L_{\updownarrow}$ is the length of the longest path. The maximum value of *Pf* is 1 and corresponds to a root system with all paths having the same length (i.e., full symmetry of the root system).

After extracting the Pf, the index of dominance of the principal axis or taproot (DA) was also calculated. DA differentiates root systems with strong central root dominance, where the taproot grows more vigorously than the branching roots, from systems with a more distributed branching pattern and no clearly dominant taproot (Martin-Ducup et al., 2020). To calculate the DA, *A1* was determined for each single path:

$$A1 = (1 - \Delta Ang) + (1 - \Delta Diam) + HP + \bar{A}n\bar{g} \tag{2}$$

where *ΔAng* and *ΔDiam* represent the average relative differences in zenithal angles and diameters, respectively, between two consecutive segments (the 'mother' and 'daughter' within the same path). *Diam* refers to the root branch diameter measured at the midpoint of each segment; *HP* is the relative length of the path, calculated as the ratio between the length of the path's largest segment and the total length of the root system; and $\bar{A}n^{-}g^{-}$ represents the mean relative zenithal angle of all segments within a path, ranging from 0 (completely horizontal) to 1 (completely vertical). The *A1* metric spans from 0 to 4, where 0 is a path with a low likelihood of being the principal axis, and 4 indicates a high likelihood (see a more detailed description for this in Martin-Ducup et al., 2020, Supplementary Material). For each root system, the path with the highest A1 value was designated as the principal axis, *PA1*. The dominance index (*DA*) of a given root system's principal axis was then defined as the difference between *PA*1 and the mean *A*1 value across all paths *i* in the root system, where *np* denotes the number of paths:

$$DA = PA1 - \frac{\sum A1i}{np} \tag{3}$$

For analyzing root system ramifications, we used the fork rate, which represents the average number of forks per meter and reflects the root system's ability to diverge from its growth pattern and spread laterally, as described by Barthelemy and Caraglio (2007) in the concept of sequential reiteration. The number of forks were calculated by examining the root QSM from the base to the top. A ramification point was classified as a fork if at least one non-dominant daughter had a diameter of at least 75 % of the largest daughter's diameter. Once a fork was identified, all retained daughter branches were traced to the next ramification point and beyond until the end of the path.

All estimations were performed using the "aRchi" package (Martin and Lecigne, 2021) in RStudio (RStudio Team, 2020) and point clouds were filtered before QSM generation.

2.4. Statistical analysis

First, to evaluate how TLS-derived root architectural metrics (Pf, DA, and fork rate) varied between revegetation methods (outplanting *versus* direct seeding), we used Non-Metric Multidimensional Scaling (NMDS) and Permutational Multivariate Analysis of Variance (PERMANOVA). NMDS was used to visualize patterns in root architecture, while PERMANOVA tested whether the observed differences between revegetation methods were statistically significant. Both NMDS and PERMANOVA were assessed based on the Bray-Curtis dissimilarity matrix with 999 permutations. To further explore these differences, we conducted separate linear models for each root metric (i.e., both TLS-derived metrics and manually measured variables) using revegetation method as the independent variable. These linear models allowed for a detailed analysis of how each revegetation method influenced individual root characteristics, enabling us to assess specific effects rather than relying solely on broader trends identified in multivariate analyses.

To assess differences in the resprouting capacity of holm oaks between outplanting and seeding, we fitted a binomial logistic model with resprouting in 2023 (alive = 1, dead = 0) as the response variable. Unlike the linear regressions used for continuous variables, this model designed for binary outcomes estimated the likelihood of survival using the logit function:

$$\log\left(\frac{p}{1-p}\right) = \beta0 + \beta1RM + \beta2L2021 + \beta3(RMxL2021) + \beta4X + \beta5Y \tag{4}$$

where *p* is the probability of resprouting success, *RM* is the revegetation method, *L2021* is the leader shoot length measured in 2021, the interaction term (*RM* × *L2021*) tests whether initial plant size influences resprouting differently between outplanting and seeding, and X and Y are spatial coordinates of each plant within the common garden site, which were used as covariates. Additionally, we ran an analogous model in which *L2021* was replaced by *S2021*, while keeping the same covariates and interaction term, in order to test the effects for shoot biomass accumulation measured in 2021:

$$\log\left(\frac{p}{1-p}\right) = \beta0 + \beta1RM + \beta2S2021 + \beta3(RMxS2021) + \beta4X + \beta5Y \tag{5}$$

Finally, we examined how the leader shoot length of resprouted plants in 2023 was affected by the revegetation method (levels: "outplanting" and "direct seeding") using another linear regression, with leader shoot length in 2021 and plant spatial position (coordinates X, Y) as covariates. As in the binomial logistic model, we included an interaction term (*revegetation method* × *leader shoot length measured in 2021)* to assess whether initial plant size influenced the leader shoot length in 2023 differently between outplanting and seeding.

All analyses were conducted in R (R Core Team, 2023), and NMDS and PERMANOVA were performed using the "vegan" package (Oksanen et al., 2022).

3. Results

3.1. Differences in root architectural metrics between outplanting and seeding

The NMDS revealed distinct patterns in TLS-derived root architectural metrics between the outplanted and seeded holm oak root systems (Fig. 2). Root systems from the seeding method were predominantly associated with metrics reflecting apical dominance (DA and Pf), while those from the outplanting method were more strongly linked to metrics indicating root ramifications (higher Fork rate) (Fig. 2). The PERMANOVA analysis revealed significant differences in root architectural metrics between the two revegetation methods (F = 6.686; p-value = 0.006).

This pattern was corroborated by the individual anlaisys of TLS-derived variables. The path fraction (Pf) was significantly higher in seeded holm oaks than in outplanted individuals, reflecting a stronger dominance of the apical axis (i.e., values closer to 1; Fig. 3, Table 1), whereas DA did not differ significantly between revegetation methods (Fig. 3, Table 1). Fork rate was significantly higher in outplanted individuals, indicating a greater number of ramifications compared to those from seeding (Fig. 3, Table 1).

Taproot diameter and mean diameter of branching roots measured manually were larger in outplanted holm oaks, averaging 3.40 ± 0.22 cm and 2.64 ± 0.14 cm, respectively, compared to 1.01 ± 0.03 cm and 0.74 ± 0.04 cm in seeded individuals, reflecting the larger size of outplanted plants. Nonetheless, root branching density was significantly higher in outplanted than in seeded plants (Fig. 4, Table 1). Additionally, the ratio of mean branching root diameter to taproot diameter was significantly greater in outplanted holm oaks than in seeded ones (Fig. 4, Table 1). Contrarily, the root:shoot ratio was significantly higher in seeded than in outplanted holm oaks (Fig. 4, Table 1).

3.2. Differences in resprouting capacity between outplanting and seeding

The binomial logistic models showed that resprouting was significantly influenced by revegetation method, leader shoot length and shoot biomass measured in 2021, and the interactions between revegetation method and leader shoot length and revegetation method and shoot biomass (Table 2). Outplanted holm oaks had a significantly lower likelihood of resprouting than those from direct seeding, with predicted probabilities of 97.6 % and 99.96 %, respectively, for individuals with average leader shoot length (Table 2). The likelihood of resprouting was also higher for holm oaks with greater leader shoot length and shoot biomass in 2021 (Table 2). Nonetheless, this effect was weaker for outplanted individuals, as indicated by a negative interaction term (Table 2). This reflects a size-dependent resprouting response that varied between revegetation methods: among smaller holm oaks (i.e., with a leader shoot length below ~25 cm), outplanted individuals had a higher probability of resprouting, whereas taller holm oaks (i.e., with a leader shoot length between 25 and 125 cm) were more likely to resprout when established by direct seeding (Fig. 5). There was also some spatial variability in resprouting likelihood, as indicated by a significant effect of the Y coordinate (Table 2).

Leader shoot length in 2023 was significantly influenced by leader shoot length in 2021 (F = 12.469, p-value < 0.001) and spatial coordinates (F = 7.303, p-value < 0.001 for X, and F = 7.732, p-value < 0.001 for Y) (Table 2). However, revegetation method had no significant effect (F = −1.026, p-value = 0.305), and its interaction with leader shoot length measured in 2021 was also non-significant (F = −1.026, p-value = 0.305), reflecting that changes in plant size followed a consistent pattern across revegetation treatments (Table 2).

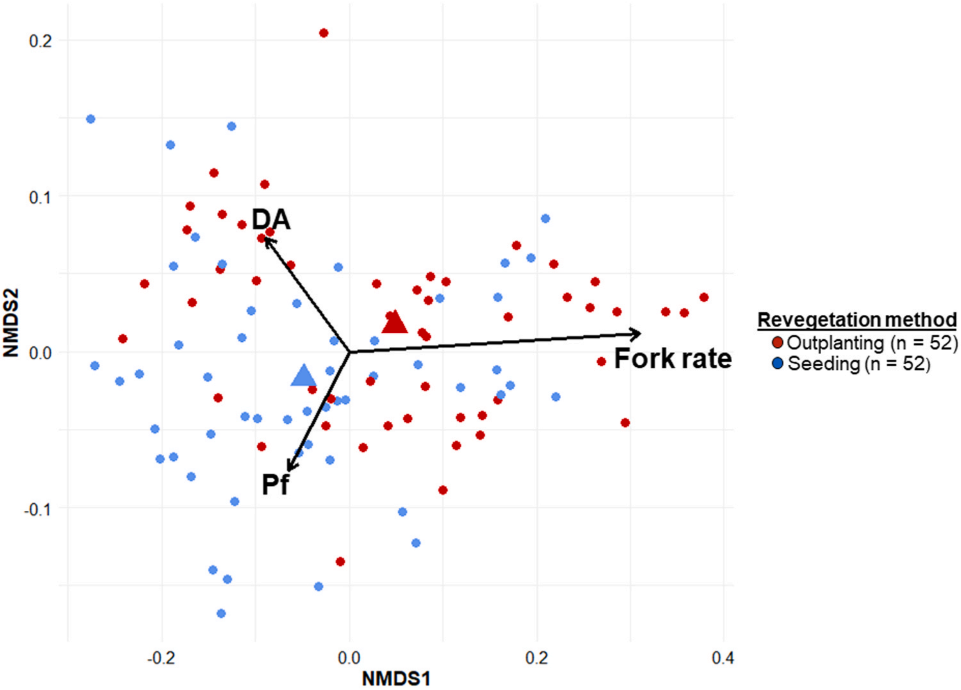

Fig. 2. Non-Metric Multidimensional Scaling (NMDS) showing the distribution of the TLS-derived root architectural metrics of the 104 samples (outplanting in red and seeding in blue). Pf: Path fraction; DA: Index of Dominance of the Principal Axis; Fork rate: Number of forks per meter. Centroids (triangles) were drawn for each revegetation method and follow the same colors as the points.

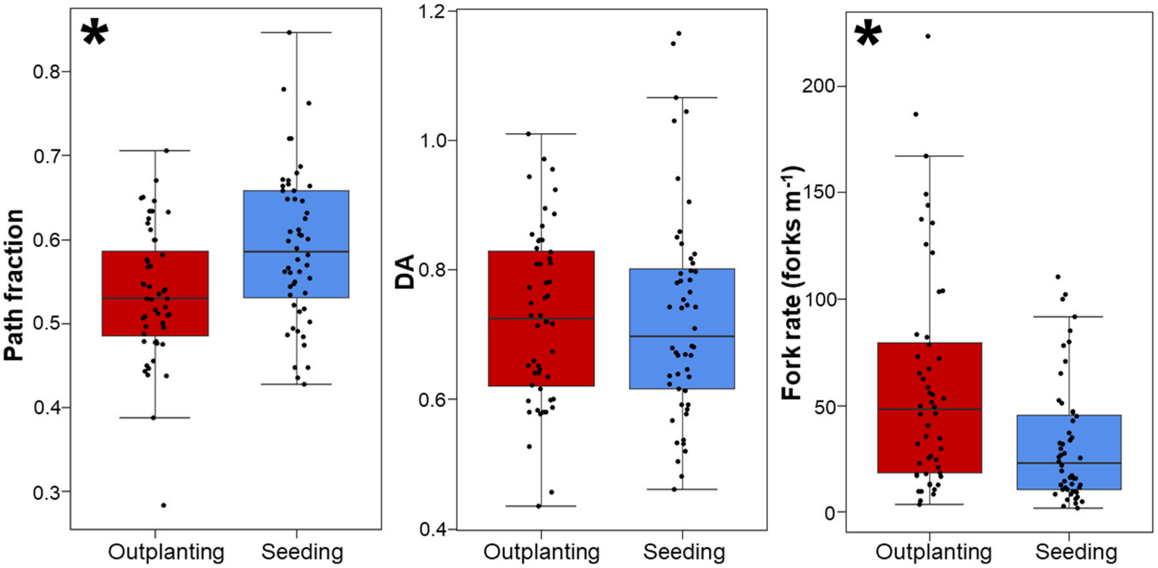

Fig. 3. Boxplots showing the values of the TLS-derived root architectural metrics (Path fraction (Pf); Index of Dominance of the Principal Axis (DA), and fork rate) within each revegetation method. Each box spans the interquartile range, whiskers extend up to 1.5 times the interquartile range, the medians are represented as black lines and black dots are the root samples (n = 52 for each revegetation method). Asterisks indicate significant differences between revegetation methods for each measured variable.

Table 1
Mean ± SE of TLS-derived and manually-measured root variables. Results of the linear models testing for differences between revegetation methods are shown, including F-values and associated p-values for each variable. Significant models (p-value < 0.05) are highlighted in bold.

	Outplanting	Seeding	F	p-value
TLS-derived variables				
Path fraction	0.53 ± 0.01	0.59 ± 0.01	**11.80**	**< 0.001**
DA	0.75 ± 0.02	0.73 ± 0.02	0.01	> 0.05
Fork rate (forks m^{-1})	59.96 ± 7.19	32.36 ± 4.03	**11.20**	**< 0.001**
Manually-measured variables				
Root branching density (branching roots m^{-1})	22.43 ± 1.53	12.35 ± 1.04	**29.75**	**< 0.001**
Ratio of mean branching root diameter to taproot diameter	0.36 ± 0.03	0.30 ± 0.02	**4.05**	**0.047**
Root:shoot ratio	0.83 ± 0.35	1.14 ± 0.62	**10.06**	**0.002**

4. Discussion

Our findings revealed that seeded holm oaks had more pronounced apical dominance and a higher root-to-shoot ratio. In contrast, outplanted individuals exhibited increased root ramification and branching density, along with thicker taproots and lateral roots, and a higher ratio between the average diameter of branching roots and that of the taproot. Unlike previous studies that have focused on young oak seedlings grown in controlled nursery or greenhouse environments (e.g., Pemán et al., 2006; Tsakaldimi et al., 2007; González-Rodríguez et al., 2011; Oliet et al., 2015), we examined plants that have been established in the field for five years, following either seedling outplanting or direct acorn seeding treatments. Furthermore, we used terrestrial laser scanning, a highly accurate technology that has previously been applied to assess aboveground tree structure (Martin-Ducup et al., 2020), for measuring root architecture for the first time. This allowed for precise, three-dimensional mapping of root systems, providing detailed information on the morphology of the coarse root system, dominance of the taproot, and root branching patterns of 4 and 5 year old trees at the sapling stage.

Our results support our first hypothesis, showing that holm oaks from the seeding treatment developed a more dominant taproot, as reflected by their higher path fraction (Fig. 6). In contrast, outplanted oaks exhibited greater root ramification, characterized by a higher fork rate and root branching density. Moreover, outplanted individuals had thicker taproots and branching roots. Interestingly, the ratio of mean branching root diameter to taproot diameter was higher in outplanted individuals than in seeded ones, which likely reflects growth constraints imposed by the nursery containers that restricted taproot development, and led to long-lasting root deformations after outplanting. This pattern reflects a trade-off where outplanted individuals, grown in constrained environments like forest trays, develop a root system with enhanced lateral branching, likely to optimize resource acquisition within their confined root space. In addition, the limited size of the containers causes the abortion of the tap root, which may further enhance the proliferation of thicker secondary roots, resulting in deformed, brush-like root systems (Mucha et al., 2018; Grossnickle and Ivetić, 2022; Fig. 6). In contrast, seeded individuals retained a well-defined taproot with comparatively few ramifications (Fig. 6). Deep taproots reduce the dependence on surface soil layers, which are more prone to drying out, and minimize the overexploitation of resources in narrow soil profiles (Pierret et al., 2016). Thus, direct seeding led to a root system better adapted to natural conditions, with vertical growth aiding both access to deep soil resources and greater stability against wind disturbances (Grossnickle and Ivetić, 2017; Löf et al., 2019). Our results are further supported by the higher root:shoot ratio observed in the seeding treatment compared to the outplanting treatment. However, this result should be interpreted with caution, as the shoot biomass of 5-year old outplanted oaks was larger than that of seeded individuals, and because the depth at which roots were extracted was similar across treatments and limited by the machinery. As a result, the root:shoot ratio may be underestimated for the seeding treatment. Nonetheless, this finding aligns with the hypothesis that direct seeding promotes better root development in holm oaks.

Our second hypothesis was partially supported: seeded holm oaks showed greater resprouting than outplanted ones, but only in the intermediate size range (25–125 cm). For larger plants, resprouting was similar regardless of the revegetation method, while smaller outplanted plants resprouted more, likely due to the initial advantages gained from controlled nursery conditions. In contrast, the stronger resprouting

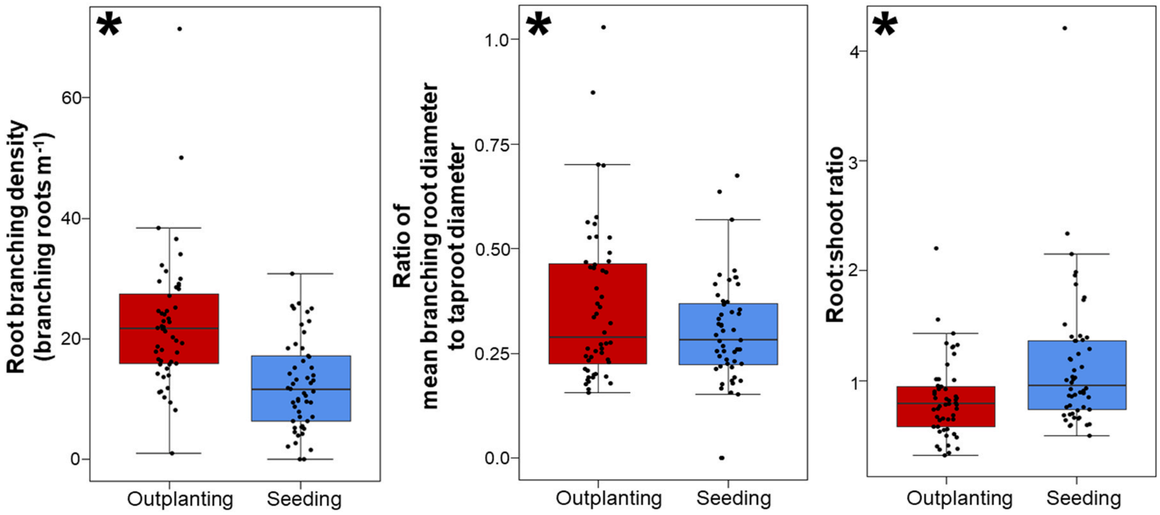

Fig. 4. Boxplots showing the values of the manually-measured variables root branching density, ratio of mean branching root diameter to taproot diameter, and root: shoot ratio within each revegetation method. Each box spans the interquartile range, whiskers extend up to 1.5 times the interquartile range, the medians are represented as black lines and black dots are the root samples (n = 52 for each revegetation method). Asterisks indicate significant differences between revegetation methods for each measured variable.

Table 2
Coefficients ± SE, z-values, and p-values from the binomial logistic model testing resprouting probability in relation to a) leader shoot length measured in 2021 and b) shoot biomass measured in 2021. Significant terms of the model ($p < 0.05$) are highlighted in bold.

	Coefficient ± SE	z	p-value
a) Leader shoot length measured in 2021			
Intercept	**5.23 ± 0.87**	**6.01**	**< 0.001**
Revegetation method (outplanting)	**−2.72 ± 0.89**	**−3.04**	**< 0.05**
Leader shoot length$_{2021}$	**5.22 ± 0.83**	**0.83**	**< 0.001**
Revegetation method (outplanting) x Leader shoot length$_{2021}$	**−2.80 ± 0.89**	**−3.14**	**< 0.05**
X	−0.16 ± 0.12	−1.33	> 0.05
Y	**0.29 ± 0.12**	**2.44**	**< 0.05**
b) Shoot biomass measured in 2021			
Intercept	**8.01 ± 1.89**	**4.25**	**< 0.001**
Revegetation method (outplanting)	**−5.20 ± 1.91**	**−2.73**	**< 0.05**
Shoot biomass$_{2021}$	**14.27 ± 3.41**	**4.19**	**< 0.001**
Revegetation method (outplanting) x Shoot biomass$_{2021}$	**−10.57 ± 3.47**	**−3.05**	**< 0.05**
X	−0.02 ± 0.11	−0.22	> 0.05
Y	**0.27 ± 0.11**	**2.52**	**< 0.05**

ability of seeded individuals in the intermediate size range likely reflects their deeper and more developed taproot, and more resource autonomy. Direct acorn seeding facilitates the development of a taproot that can extend beyond 1 m in depth within the first year of growth (Löf and Welander, 2004; Zadworny et al., 2021). This deep taproot not only promotes vertical growth but also supports the production of ephemeral fine roots in deeper soil layers, enhancing nutrient and water absorption, and reducing competition for water resources with close shrubs and grasses mainly exploiting soil surface resources (Kościelniak et al., 2021). In contrast, outplanted holm oaks, with their shallower root systems, may be less efficient at resource acquisition, limiting their ability to produce new shoots and recover following disturbances (Grossnickle, 2005). Interestingly, the age difference between treatments (5 years for outplanting and 4 for seeding) turns out to make our results more conservative, as the advantage of seeding over planting in terms of resprouting ability would most likely have increased if seeding had occurred one year earlier. Thus, direct acorn seeding may enhance oak resilience, favoring the establishment of mature and healthy trees with greater stability (Grossnickle, 2018; Löf et al., 2019; Castro et al., 2021; Navarro et al., 2023).

The high inherent variability of soils in Mediterranean ecosystems, where resource availability is often patchy (Sardans and Peñuelas, 2013), underscores the importance of choosing an appropriate revegetation method for species like holm oaks. Soil heterogeneity influences root development and requires plants to adjust their rooting strategies to efficiently capture spatially uneven and limited resources (de la Riva et al., 2018; Marañón et al., 2020). Traits such as root diameter and root branching density, which were measured in this study, are shaped by fine-scale soil conditions to optimize resource acquisition (Bardgett et al., 2014; Freschet et al., 2021). Our results suggest that direct acorn seeding may offer an advantage for acclimation to heterogeneous local conditions. As acorns germinate and grow directly in the field, they produce a deep and vertically-oriented taproot able to respond to nutrient or moisture gradients and generate fine roots for optimal resource absorption where needed (López et al., 2001; Serrada et al., 2017). In addition, the enhanced resprouting capacity of seeded holm oaks may further improve their survival and establishment in patchy soils and their resilience against disturbances such as browsing. Contrarily, seedlings grown in controlled nursery environments with uniform substrates and regulated watering present limitations in taproot depth, that limits the capacity of the plant to explore and acclimate effectively to soil variability (Pemán et al., 2017). This highlights an important trade-off: while nursery-grown seedlings may initially establish more reliably due to previous nursery controlled conditions, their restricted root architecture may limit their ability to adjust to heterogeneous environmental conditions once outplanted. It should be noted, however, that in the outplanting treatment, the seedlings were grown in 300 ml containers, a size commonly used in oak nurseries in the Mediterranean Basin but still considered small. Larger containers, particularly taller ones, could help reduce root deformities and improve outplanting performance. Future studies should also consider the economic and logistical trade-offs associated with the use of larger container volumes.

Our findings are particularly relevant for the restoration of Mediterranean open woodlands and forests, where drought is driving widespread tree mortality and root system damage (Ruiz-Gómez et al., 2019; Encinas-Valero et al., 2022). Direct acorn seeding can enhance holm oak resilience by fostering deeper root systems that access water from lower soil layers. However, its success may be limited by early-stage drought and seed predation (Leverkus et al., 2015a,b; Löf et al., 2019).

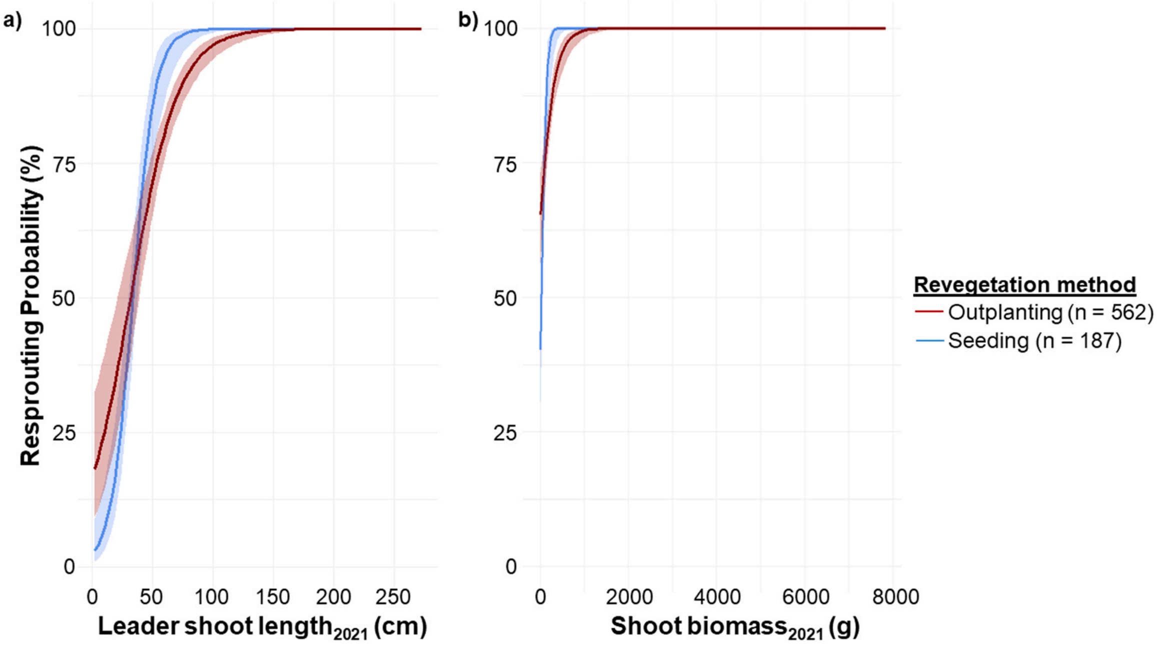

Fig. 5. Resprouting probability of outplanted and seeded holm oaks in relation to a) the leader shoot length measured in 2021, and b) the shoot biomass measured in 2021.

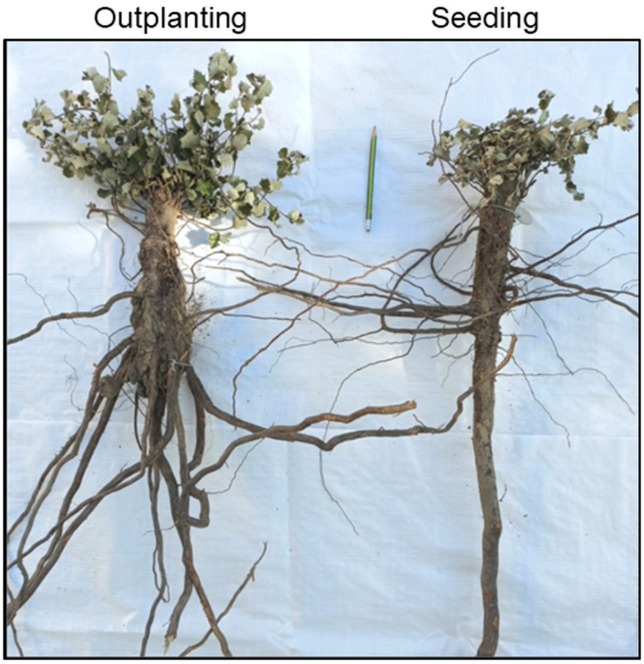

Fig. 6. Examples of root systems of one outplanted individual (left) and one seeded individual (right). The aboveground part of the plants corresponds to the resprouting shoots in May 2022, ca. six months after cutting of the aerial part (see Material and Methods for details). Note the strong development of the taproot in the seeded treatment.

Integrating precision forestry techniques, such as the strategic placement of seeds in microsites with favorable abiotic conditions and shelter, can help overcome these challenges (Castro et al., 2021). Altogether, direct acorn seeding should be considered a key revegetation strategy to improve early establishment and support the mid-term stability of holm oak ecosystems under escalating drought stress in Mediterranean landscapes.

CRediT authorship contribution statement

Alexandro B. Leverkus: Conceptualization, Field work, Writing - review & editing. **María Noelia Jiménez:** Field work. **Domingo Alcaraz-Segura:** Conceptualization, Methodology. **Marino Pedro Reyes-Martín:** Field work, Data curation. **Emilio Rodríguez-Caballero:** Methodology, Writing - review & editting, Data curation. **Francisco B. Navarro:** Conceptualization, Field work, Funding acquisition. **Jorge Castro:** Conceptualization, Methodology, Field work, Writting - review & editing, Funding acquisition. **Raquel Juan-Ovejero:** Field work, Writing - original draft, Visualization, Formal analysis, Data curation.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgements

This study was supported by the projects AIDroneSeeding (Ref. PID2023–147483OB-I00), funded by MICIU/AEI/10.13039/501100011033, and RESISTE (Ref. P18-RT-1927), funded by the Consejería de Transformación Económica, Industria, Conocimiento y Universidades, Junta de Andalucía. Projects TED2021–130976B-I00 and TED2021–132332B-C21 funded by MICIU/AEI/10.13039/501100011033 and by the European Union Next GenerationEU/PRTR; RESISTRES (C-EXP267-UGR23) from University of Granada/ Junta de Andalucía/ FEDER, and SMART-OAK (funded by the EU through FORWARDS (Horizon Europe Project No. 101084481) grants to third parties managed by European Forest Institute) are also acknowledged. Thanks to the IFAPA projects AVA201601.19 (NUTERA-DE) and AVA2019.004 (NUTERA-DE II), co-financed (80 %) by the EU FEDER program and the Andalusian Government, was possible to establish and maintain this experiment in the early stages. E. Rodríguez-Caballero was supported by the Ramón y Cajal fellowship (RYC2020–030762-I). R. Juan-Ovejero holds a postdoctoral contract (Ref. ED481B-2022–006) funded by the Consellería de Cultura, Educación, Formación Profesional e

Universidades from the Xunta de Galicia.

Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.foreco.2025.123187.

Data availability

I have shared the link to my data at the Attach File step.

Effect of revegetation method (seedling outplanting versus direct seeding) on holm oak root architecture: implications for restoration success under a global change scenario - DATASET (Mendeley Data)

References

Austin, K.G., Baker, J.S., Sohngen, B.L., Wade, C.M., Daigneault, A., Ohrel, S.B., Bean, A., 2020. The economic costs of planting, preserving, and managing the world's forests to mitigate climate change. Nat. Commun. 11 (1), 5946.

Barbeta, A., Peñuelas, J., 2017. Increasing carbon discrimination rates and depth of water uptake favor the growth of Mediterranean evergreen trees in the ecotone with temperate deciduous forests. Glob. Change Biol. 23 (12), 5054–5068.

Bardgett, R.D., Mommer, L., De Vries, F.T., 2014. Going underground: root traits as drivers of ecosystem processes. Trends Ecol. Evol. 29 (12), 692–699.

Barthelemy, D., Caraglio, Y., 2007. Plant architecture: a dynamic, multilevel and comprehensive approach to plant form, structure and ontogeny. Ann. Bot. 99, 375–407.

Bastin, J.F., Finegold, Y., Garcia, C., Mollicone, D., Rezende, M., Routh, D., Crowther, T. W., 2019. The global tree restoration potential. Science 365 (6448), 76–79.

Castro, J., Leverkus, A.B., Fuster, F., 2015. A new device to foster oak forest restoration via seed sowing. N. For. 46, 919–929.

Castro, J., Morales-Rueda, F., Navarro, F.B., Löf, M., Vacchiano, G., Alcaraz-Segura, D., 2021. Precision restoration: a necessary approach to foster forest recovery in the 21st century. Restor. Ecol. 29 (7), e13421.

Ceccon, E., González, E.J., Martorell, C., 2016. Is direct seeding a biologically viable strategy for restoring forest ecosystems? Evidences from a Meta-analysis. Land Degrad. Dev. 27 (3), 511–520.

Cernansky, R., 2018. How to rebuild a forest. Nature 560 (7720), 542–544.

Chirino, E., Vilagrosa, A., Hernández, E.I., Matos, A., Vallejo, V.R., 2008. Effects of a deep container on morpho-functional characteristics and root colonization in *quercus suber* L. Seedlings for reforestation in Mediterranean climate. For. Ecol. Manag. 256 (5), 779–785.

Cook-Patton, S.C., Drever, C.R., Griscom, B.W., Hamrick, K., Hardman, H., Kroeger, T., Ellis, P.W., 2021. Protect, manage and then restore lands for climate mitigation. Nat. Clim. Change 11 (12), 1027–1034.

Dey, D.C., Jacobs, D., McNabb, K., Miller, G., Baldwin, V., Foster, G., 2008. Artificial regeneration of major oak (*quercus*) species in the eastern United States—A review of the literature. For. Sci. 54, 77–106.

Encinas-Valero, M., Esteban, R., Hereş, A.M., Vivas, M., Fakhet, D., Aranjuelo, I., Curiel Yuste, J., 2022. Holm oak decline is determined by shifts in fine root phenotypic plasticity in response to belowground stress. N. Phytol. 235 (6), 2237–2251.

Espelta, J.M., Retana, J., Habrouk, A., 2003. Resprouting patterns after fire and response to stool cleaning of two coexisting Mediterranean oaks with contrasting leaf habits on two different sites. For. Ecol. Manag. 179 (1-3), 401–414.

Fernández-Habas, J., Fernández-Rebollo, P., Casado, M.R., Moreno, A.M.G., Abellanas, B., 2019. Spatio-temporal analysis of oak decline process in open woodlands: a case study in SW Spain. J. Environ. Manag. 248, 109308.

Freschet, G.T., Pagès, L., Iversen, C.M., Comas, L.H., Rewald, B., Roumet, C., McCormack, M.L., 2021. A starting guide to root ecology: strengthening ecological concepts and standardising root classification, sampling, processing and trait measurements. N. Phytol. 232 (3), 973–1122.

Gil-Pelegrín, E., Peguero-Pina, J.J., Sancho-Knapik, D. (Eds.), 2017. Oaks physiological ecology: Exploring the functional diversity of genus Quercus L. Springer.

González-Rodríguez, V., Navarro-Cerrillo, R.M., Villar, R., 2011. Artificial regeneration with *quercus ilex* L. And *quercus suber* L. By direct seeding and planting in Southern Spain. Ann. For. Sci. 68, 637–646.

Grossnickle, S., 2018. Seedling establishment on a forest restoration site: an ecophysiological perspective. Reforesta (6), 110–139.

Grossnickle, S.C., 2005. Importance of root growth in overcoming planting stress. N. For. 30 (2), 273–294.

Grossnickle, S.C., Ivetić, V., 2017. Direct seeding in reforestation–a field performance review. Reforesta (4), 94–142.

Grossnickle, S.C., Ivetić, V., 2022. Root system development and field establishment: effect of seedling quality. N. For. 53 (6), 1021–1067.

Hua, F., Bruijnzeel, L.A., Meli, P., Martin, P.A., Zhang, J., Nakagawa, S., Balmford, A., 2022. The biodiversity and ecosystem service contributions and trade-offs of forest restoration approaches. Science 376 (6595), 839–844.

Juan-Ovejero, R., Castro, J., Navarro, F.B., Moreno-Rojas, J.M., Jimenez, M.N., Leverkus, A.B., Querejeta, J.I., 2024. Large physiological plasticity of water-and nutrient-use traits in *quercus ilex* L. Within and across populations: implications for Mediterranean forest persistence under global change. Environ. Exp. Bot. 228, 106053.

Keller, A.A., Fox, J., 2019. Giving credit to reforestation for water quality benefits. PLoS One 14 (6), e0217756.

Kościelniak, P., Glazińska, P., Kęsy, J., Zadworny, M., 2021. Formation and development of taproots in deciduous tree species. Front. Plant Sci. 12, 772567.

Laestadius, L., Maginnis, S., Minnemeyer, S., Potapoy, P., Saint-Laurent, C., Sizer, N., 2011. Mapping opportunities for forest landscape restoration. Unasylva 62 (238), 47–48.

Lázaro-González, A., Andivia, E., Hampe, A., Hasegawa, S., Marzano, R., Santos, A.M., Leverkus, A.B., 2023. Revegetation through seeding or planting: a worldwide systematic map. J. Environ. Manag. 337, 117713.

Leverkus, A.B., Castro, J., Delgado-Capel, M.J., Molinas-González, C., Pulgar, M., Marañón-Jiménez, S., Querejeta, J.I., 2015b. Restoring for the present or restoring for the future: enhanced performance of two sympatric oaks (*quercus ilex* and *quercus pyrenaica*) above the current forest limit. Restor. Ecol. 23 (6), 936–946.

Leverkus, A.B., Rojo, M., Castro, J., 2015a. Habitat complexity and individual acorn protectors enhance the post-fire restoration of oak forests via seed sowing. Ecol. Eng. 83, 276–280.

Lewis, S.L., Wheeler, C.E., Mitchard, E.T., Koch, A., 2019. Restoring natural forests is the best way to remove atmospheric carbon. Nature 568 (7750), 25–28.

Löf, M., Welander, N.T., 2004. Influence of herbaceous competitors on early growth in direct seeded *fagus sylvatica* L. And *quercus robur* l. Ann. For. Sci. 61 (8), 781–788.

Löf, M., Castro, J., Engman, M., Leverkus, A.B., Madsen, P., Reque, J.A., Gardiner, E.S., 2019. Tamm review: direct seeding to restore oak (*quercus* spp.) forests and woodlands. For. Ecol. Manag. 448, 474–489.

López, B., Sabaté, S., Gracia, C.A., 2001. Vertical distribution of fine root density, length density, area index and mean diameter in a *quercus ilex* forest. Tree Physiol. 21 (8), 555–560.

Mansourian, S., Derkyi, M., Djenontin, I., Elias, M., Oldekop, J., Pacheco, P., Burns, J., Diederichsen, A., Kleine, M., Vallauri, D., Walder, B., 2024. Human dimensions of forest landscape restoration. IUFRO, Vienna, p. 76.

Mansourian, S., Adams, W.M., Bouazza, K., Ferreira, J.N., Ganz, D.J., Hurd, J., Kull, C.A., 2025. Reconciling global and local benefits of forest restoration: a shared interdisciplinary perspective. For. Policy Econ. 170, 103381.

Marañón, T., Navarro-Fernández, C.M., Gil-Martínez, M., Domínguez, M.T., Madejón, P., Villar, R., 2020. Variation in morphological and chemical traits of Mediterranean tree roots: linkage with leaf traits and soil conditions. Plant Soil 449, 389–403.

Mariotti, B., Maltoni, A., Jacobs, D.F., Tani, A., 2015. Container effects on growth and biomass allocation in *quercus robur* and *juglans regia* seedlings. Scand. J. For. Res. 30 (5), 401–415.

Martin, O., & Lecigne, B. (2021). aRchi: Quantitative Structural Model ("QSM") Treatment for Tree Architecture. R package version 2.1.3, ⟨https://cran.r-project.org/web/packages/aRchi⟩.

Martin-Ducup, O., Ploton, P., Barbier, N., Momo Takoudjou, S., Mofack II, G., Kamdem, N.G., Fourcaud, T., Sonké, B., Couteron, P., Pélissier, R., 2020. Terrestrial laser scanning reveals convergence of tree architecture with increasingly dominant crown canopy position. Funct. Ecol. 34 (12), 2442–2452.

Martín-Sánchez, R., Peguero-Pina, J.J., Alonso-Forn, D., Ferrio, J.P., Sancho-Knapik, D., Gil-Pelegrín, E., 2022. Summer and winter can equally stress holm oak (*quercus ilex* L.) in Mediterranean areas: a physiological view. Flora 290, 152058.

Ménard, I., Thiffault, E., Kurz, W.A., Boucher, J.F., 2023. Carbon sequestration and emission mitigation potential of afforestation and reforestation of unproductive territories. N. For. 54 (6), 1013–1035.

Mucha, J., Jagodziński, A.M., Bułaj, B., Łakomy, P., Talaśka, A.M., Oleksyn, J., Zadworny, M., 2018. Functional response of *quercus robur* L. To taproot pruning: a 5-year case study. Ann. For. Sci. 75, 1–12.

Navarro, F.B., Caño, A.B., Gálvez, C., Kazani, A., Carbonero, M.D., Jiménez, M.N., 2023. Key factors in direct acorn seeding for the successful restoration of open oak woodlands. For. Ecol. Manag. 546, 121314.

Navarro-Cerrillo, R.M., Gómez, F.J.R., Cabrera-Puerto, R.J., Sánchez-Cuesta, R., Rodriguez, G.P., Pérez, J.L.Q., 2018. Growth and physiological sapling responses of eleven *quercus ilex* ecotypes under identical environmental conditions. For. Ecol. Manag. 415, 58–69.

Oksanen, J., Simpson, G., Blanchet, F., Kindt, R., Legendre, P., Minchin, P., O'Hara, R., Solymos, P., Stevens, M., Szoecs, E., Wagner, H., Barbour, M., Bedward, M., Bolker, B., Borcard, D., Carvalho, G., Chirico, M., De Caceres, M., Durand, S., Evangelista, H., FitzJohn, R., Friendly, M., Furneaux, B., Hannigan, G., Hill, M., Lahti, L., McGlinn, D., Ouellette, M., Ribeiro Cunha, E., Smith, T., Stier, A., Ter Braak, C., & Weedon, J. (2022). Vegan: Community ecology package. R package version 2.6-4. ⟨https://cran.r-project.org/package=vegan⟩.

Oliet, J.A., Vázquez de Castro, A., Puértolas, J., 2015. Establishing *quercus ilex* under Mediterranean dry conditions: sowing recalcitrant acorns versus planting seedlings at different depths and tube shelter light transmissions. N. For. 46, 869–883.

Osonubi, O., Davies, W.J., 1981. Root growth and water relations of oak and birch seedlings. Oecologia 51, 343–350.

Padilla, F.M., Pugnaire, F.I., 2007. Rooting depth and soil moisture control Mediterranean woody seedling survival during drought. Funct. Ecol. 21 (3), 489–495.

Pausas, J.G., Keeley, J.E., 2014. Evolutionary ecology of resprouting and seeding in fire-prone ecosystems. N. Phytol. 204 (1), 55–65.

Pemán, J., Voltas, J., Gil-Pelegrín, E., 2006. Morphological and functional variability in the root system of *quercus ilex* L. Subject to confinement: consequences for afforestation. Ann. For. Sci. 63, 425–430.

Pemán, J., Chirino, E., Espelta, J.M., Jacobs, D.F., Martín-Gómez, P., Navarro Cerrillo, R., Oliet, J.A., Vilagrosa, A., Villar-Salvador, P., Gil-Pelegrín, E., 2017. Physiological keys for natural and artificial regeneration of oaks (2017). In: Gil-Pelegrín, E., Peguero-Pina, J.J., Sancho-Knapik, D. (Eds.), Oaks physiological ecology: Exploring the functional diversity of genus Quercus L. Springer, pp. 453–513.

Pierret, A., Maeght, J.L., Clément, C., Montoroi, J.P., Hartmann, C., Gonkhamdee, S., 2016. Understanding deep roots and their functions in ecosystems: an advocacy for more unconventional research. Ann. Bot. 118 (4), 621–635.

Preece, N.D., van Oosterzee, P., Lawes, M.J., 2023. Reforestation success can be enhanced by improving tree planting methods. J. Environ. Manag. 336, 117645.

Pyttel, P.L., Fischer, U.F., Suchomel, C., Gärtner, S.M., Bauhus, J., 2013. The effect of harvesting on stump mortality and re-sprouting in aged oak coppice forests. For. Ecol. Manag. 289, 18–27.

R Core Team, 2023. R: a language and environment for statistical computing. R foundation for statistical computing, Vienna, Austria. ⟨http://www.R-project.org⟩.

de la Riva, E.G., Marañón, T., Pérez-Ramos, I.M., Navarro-Fernández, C.M., Olmo, M., Villar, R., 2018. Root traits across environmental gradients in Mediterranean woody communities: are they aligned along the root economics spectrum? Plant Soil 424, 35–48.

RStudio Team, 2020. RStudio: integrated development for R. Rstudio. PBC, Boston, MA. ⟨http://www.rstudio.com⟩.

Ruiz-Gómez, F.J., Pérez-de-Luque, A., Navarro-Cerrillo, R.M., 2019. The involvement of phytophthora root rot and drought stress in holm oak decline: from ecophysiology to microbiome influence. Curr. For. Rep. 5 (4), 251–266.

Sakai, A., Sakai, S., Akiyama, F., 1997. Do sprouting tree species on erosion-prone sites carry large reserves of resources? Ann. Bot. 79 (6), 625–630.

Sardans, J., Peñuelas, J., 2013. Plant-soil interactions in Mediterranean forest and shrublands: impacts of climatic change. Plant Soil 365, 1–33.

Scotton, M., Andreatta, D., 2021. Anti-erosion rehabilitation: effects of revegetation method and site traits on introduced and native plant cover and richness. Sci. Total Environ. 776, 145915.

Serrada, R., Gómez-Sanz, V., Aroca, M.J., Otero, J., Bravo-Fernández, J.A., Roig, S., 2017. Decline in holm oak coppices (*quercus ilex* L. subsp. *ballota* (Desf.) Samp.): biometric and physiological interpretations. For. Syst. 26 (2) e06S-e06S.

Smith, D.D., Sperry, J.S., Enquist, B.J., Savage, V.M., McCulloh, K.A., Bentley, L.P., 2014. Deviation from symmetrically self-similar branching in trees predicts altered hydraulics, mechanics, light interception and metabolic scaling. N. Phytol. 201, 217–229.

Trabucco, A., Zomer, R.J., Bossio, D.A., van Straaten, O., Verchot, L.V., 2008. Climate change mitigation through afforestation/reforestation: a global analysis of hydrologic impacts with four case studies. Agric. Ecosyst. Environ. 126 (1-2), 81–97.

Tsakaldimi, M., Tsitsoni, T., Ganatsas, P., Zagas, T., 2009. A comparison of root architecture and shoot morphology between naturally regenerated and container-grown seedlings of *quercus ilex*. Plant Soil 324 (1–2), 103–113.

Walker, W.S., Gorelik, S.R., Cook-Patton, S.C., Baccini, A., Farina, M.K., Solvik, K.K., Griscom, B.W., 2022. The global potential for increased storage of carbon on land. Proc. Natl. Acad. Sci. 119 (23), e2111312119.

Wilson, E.R., Vitols, K.C., Park, A., 2007. Root characteristics and growth potential of container and bare-root seedlings of red oak (*quercus rubra* L.) in ontario, Canada. N. For. 34 (2), 163–176.

Zadworny, M., Jagodziński, A.M., Łakomy, P., Ufnalski, K., Oleksyn, J., 2014. The silent shareholder in deterioration of oak growth: common planting practices affect the long-term response of oaks to periodic drought. For. Ecol. Manag. 318, 133–141.

Zadworny, M., Jagodziński, A.M., Łakomy, P., Mucha, J., Oleksyn, J., Rodríguez-Calcerrada, J., Ufnalski, K., 2019. Regeneration origin affects radial growth patterns preceding oak decline and death–insights from tree-ring δ13C and δ18O. Agric. For. Meteorol. 278, 107685.

Zadworny, M., Mucha, J., Jagodziński, A.M., Kościelniak, P., Łakomy, P., Modrzejewski, M., Rodríguez-Calcerrada, J., 2021. Seedling regeneration techniques affect root systems and the response of quercus robur seedlings to water shortages. For. Ecol. Manag. 479, 118552.