ISSN 1021-4437, Russian Journal of Plant Physiology, 2023, Vol. 70:41. © Pleiades Publishing, Ltd., 2023.

RESEARCH PAPERS

Seed Germination and Seedling Establishment in *Pistacia atlantica* Desf. and *Pistacia lentiscus* L. under Drought

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Received September 27, 2022; revised November 23, 2022; accepted November 23, 2022

Abstract—The rehabilitation of degraded Mediterranean areas requires reintroduction of key-stone woody species. However, seed germination and seedling establishment for native species are poor. Germination behavior of *Pistacia atlantica* Desf. and *Pistacia lentiscus* L. in response to temperature (15, 20, 25 and 30°C), salinity (0, 10, 25 and 50 mM NaCl) and scarification (seeds with/without pulp) were analyzed. The response of seedling establishment to water deficit was related to soil moisture at 100 and 50% of the field capacity. Scarification facilitated germination while salinity should not exceed 50 mM NaCl and temperature must be between 20 and 25°C for *P. lentiscus* and 25°C for *P. atlantica*. In *P. atlantica*, seedling establishment was susceptible to water deficit and it showed high-water requirement. The survival of stressed *P. atlantica* seedlings are attributed to the reduction in stomatal conductance, carbon assimilation and shoot growth, and acceleration of old leaf senescence and root growth. The divergence within both species in seed germination, carbon assimilation, growth and biomass allocation are interpreted as a plasticity response, indicating a specialization to either original or resource habitats. For this reason, in reforestation programs, poor performance of planted seedlings is also associated with soil water deficit.

Keywords: *Pistacia atlantica*, *Pistacia lentiscus*, pistachio, germination, photosynthesis, temperature, drought stress

DOI: 10.1134/S1021443722602920

INTRODUCTION

In Tunisia, the *Pistacia* genus includes, with the exception of the species *Pistacia vera* L. that produces edible fruits, a species presenting other agronomic and ecological interests. However, they are endangered and threatened, with a gradual disappearance. *P. atlantica* [1], a wild endangered species from the *Anacardiaceae* family, is a dioecious tree, with semi-evergreen leaves, a drupe fruit and an extensive root system. In Tunisia, *P. atlantica* exists as isolated aged trees in arid and semi-arid areas. The multiple uses of this species can explain its reduction in several areas [2]. Despite its adaptation to hostile environments such as salinity [3], drought and nematode tolerance and its good performance as rootstock compared to *P. vera*, it is—nowadays—rarely used as rootstock for pistachio varieties [4]. *Pistacia lentiscus* is also an Anacardiaceae tree. This evergreen plant is a sclerophyllous dioecious shrub, sometimes attaining a tree growth form in more humid and protected sites. Fruits are a drupe, starting red and maturing to a glossy black color. *Pistacia lentiscus* is an endemic shrub of the Mediterranean basin [5]. *P. atlantica* and *P. lentiscus* species are multipurpose trees, useful in soil conservation [6], as rootstock for *P. vera* [4], and in industrial and medicinal applications [7]. *P. atlantica* is a tree par excellence in steppe environments. However, it can penetrate deeply into the Saharan regions. Moreover, among other species, it is more tolerant to drought [8, 9] and salinity [10]. Seed germination is a critical phase in arid and semi-arid lands and variation in germination rates has been interpreted as an adaptation to ecological conditions [5, 11, 12]. Indeed, the extinction of species in these areas can be attributed not only to major environmental factors such as drought, soil erosion and salinity, but also to the tegumentary dormancy of seeds [12]. The main limiting factors for

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Abbreviations: *G*—percentage of seed germination; *D*—dormant seeds; *FC*—field capacity; g_s—stomatal conductance; *A*—net photosynthesis rate; *E*—rate of transpiration; *A*/*E*—ratio of photosynthesis and transpiration; RWC—relative water content; L_A—leaf area; R_L—root length; R_M—roots mass; S_M—shoots dry mass; S_M/R_M—ratio between shoots dry mass and roots mass.

seeds germination are water, soil temperature and salinity. Decreased osmotic potentials cause a reduction in germination percentage and seedling growth for many species [5, 13].

Plant establishment is a critical phase of the rehabilitation of fragile ecosystems when plants are vulnerable to abiotic and biotic stresses. Climatic conditions such as high temperature and low relative humidity, which generate considerable water losses through evapotranspiration, are among common forms of stress. Spring and summer, the two major seasons in the Mediterranean region when water stress occurs, cause relatively high water pressures. The poor performance of planted seedlings is often associated with transplant shock, competing vegetation, poor soils or lack of site preparation and soil water deficit [5]. The extinction of species under prolonged Mediterranean drought is attributed to difficulties in seedling establishment. Soil moisture is critical for seedlings to establish quickly after planting [5, 14]. Some species develop morphological and physiological changes, such as leaf area reduction, increased root growth and stomatal control, improving their survival under drought [8, 15, 16]. Water stress reduces metabolic capacity and stomatal conductance, causing a decrease in photosynthesis and growth rate [15, 17] and changes in dry matter allocation [18, 19]. Drought also increases leaf senescence and causes greater biomass allocation to roots [14, 19, 20]. The initial morphological characteristics that successfully predict field performance under dry Mediterranean conditions in seedlings vary according to species. High photosynthesis, extension of roots and leaf growth are some of the major traits related to successful plant establishment [21]. Seedlings with high net photosynthesis rates and adequate soil moisture and without vegetative competition are established rapidly [14]. In addition, species from favorable productive habitats present high specific leaf area, high relative growth rate and high allocation of biomass to shoots [16, 22]. This is explained by the optimal partitioning theory, which states that plants adapt to stressful environmental conditions by changing carbon allocation to the organs collecting the most limiting resource, a form of plasticity conducive to growth maximization [8, 23]. Therefore, many morphological characteristics, such as seedling diameter and shoot height, are correlated to field success for a species [14]. In an unproductive habitat with poor nutrient availability, species with low specific leaf area and relative growth rate seem to be allocating more biomass to roots to avoid water deficits. Hence, root morphology is an effective indicator of seedling performance under water-limiting conditions [5, 14, 16, 20]. The survival of *P. lentiscus* seedlings is positively correlated to the initial seedling root collar diameter and total dry weight [24].

The rehabilitation of degraded Mediterranean areas often requires the reintroduction of keystone woody species but the establishment of seedlings of native species is frequently poor. This is partly due to insufficient knowledge of the ecology of these species at the seedling stage [5, 25]. Early mortality of seedlings is a major risk in the successful establishment of plantations in semi-arid and arid areas. Therefore, it is important to study how these seedlings adapt to water deficit. Accurate knowledge of germination requirements and seedling establishment of *P. atlantica* and *P. lentiscus* are requisite firstly to protect both endangered species and secondly to successfully use rehabilitation and rangeland restoration programs [5, 16, 25, 26].

Oilseeds for both species are characterized by low germination. This study aims to analyze the effects of pulp, temperature and salinity on seed germination in *P. atlantica* and *P. lentiscus*. Furthermore, the response of seedlings to water deficit is evaluated on the basis of the assimilation of CO_2, growth and the allocation of biomass at early seedling establishment [5, 14, 21].

MATERIALS AND METHODS

First Experiments of Germination

They were carried out at the Faculty of Sciences of Gafsa (University of Gafsa, Tunisia) on *Pistacia atlantica* seeds from *Sidi Bouzid* (Tunisia) and *Pistacia lentiscus* seeds from *Bizerte* (Tunisia). The climatic conditions are arid and humid, respectively, at these sites of seed origin. The seeds come from the trees distributed naturally in both species. The tests were conducted in plant growth chambers that allow control of temperature and light. Seeds, placed in Petri dishes, were watered three times a week. The emergence of the radicle was taken as a sign of germination. The counting of germinated seeds was performed daily.

The percentage of seed germination (*G*) was calculated according to the following formula: $G\%$ = (Number of germinated seeds)/(Number of germinated seeds opened for each repetition – number of empty seeds) × 100.

The period of rest (*D*) of a homogeneous group of seeds was given as the number of days necessary to observe the first germinated seeds. The seeds harvested directly from the *P. atlantica* and *P. lentiscus* trees were germinated without any treatment (Mechanical or chemical, except sterilization with bleach) and performing a mechanical pulping for others. Then, they were placed at room temperature between 25 and 27°C under natural lighting. To investigate seed germination with pulp and without pulp, 100 seeds for each treatment were tested (6 repetitions). The monitoring was carried out daily over a period of 45 days. The values were used to calculate the percentage of germinated seeds (*G*) and dormant ones (*D*).

Germination

It was carried out in incubators set at 15, 20, 25 and 30°C. *P. atlantica* and *P. lentiscus* seeds harvested directly from the trees (with pulp) were placed on filter paper (Whatman) in Petri dishes (100 seeds per box) and moistened with distilled water. For each treatment, five Petri dishes were used to monitor the germination. This test was used to estimate the percentage of germinated (*G*) and dormant seeds (*D*). The *P. atlantica* and *P. lentiscus* seeds (with pulp) were tested for salinity sensitivity at different concentrations of NaCl (0, 10, 25 and 50 mM), commonly observed in Tunisian soils in arid areas. For each treatment, 100 seeds were tested in Petri dishes. The test was conducted at the optimum temperature (25–27°C) and the calculated parameters were the percentage of germinated (*G*) and dormant seeds (*D*).

Second Experiment with Seedlings of *P. atlantica* and *P. lentiscus*

The seedlings were obtained from the germination process in the first trial (control seeds) and were transplanted into 3 L pots. The test was carried out at the seedling stage for the two species and subjected to two different water regimes. Pots into which seedlings were transplanted contained a mixture of sand and clay soil (2/3 : 1/3, w/w). The seedlings were grown at 27°C on a photoperiod of 12 h. At 4 months after transplantation, an irrigated treatment was applied where the soil is maintained at 100% of field capacity (*FC*) (control, well irrigated) and a stressed treatment was performed where the soil is maintained at 50% *FC* (drought). The evaporated water was returned to the plants every two days determining the weight difference on the day of water replacement versus the previous two days. The morphophysiological parameters were taken arbitrarily and were measured for 45 days (stage 1) and 100 days (stage 2) during the treatment period. Six plants were used to measure this parameter for each treatment.

The relative water content (RWC) was estimated as below:

$$\mathrm{RWC} = [(\mathrm{FW} - \mathrm{DW})/(\mathrm{TW} - \mathrm{DW})] \times 100,$$

where FW is the fresh weight of the leaf, TW is the weight at full turgidity, measured after floating the leaf for 24 h in distilled water, and DW is the weight after drying the leaf at 70°C for 48 h.

The following parameters were measured using a portable photosynthesis meter, LCi pro$^+$ (ADC Bio-Scientific Ltd). The system records the values of the net photosynthesis rate (A, μmol CO_2/m^2 s), stomatal conductance (g_s, mol H_2O/m^2 s), the transpiration rate (E, mmol H_2O/m^2 s). A/E was calculated. The number of leaves was assessed by manual counting. The measurements were taken at midday on young leaves, generally the fourth leaf after the emergent leaf.

Leaf area (L_A, cm^2 per plant) was measured twice for each seedling after the scan of the leaves by using image processing software, Mesurim pro 8 (Version 3.2). The height of shoots was measured every 15 days. After each cut, the roots were separated and their lengths measured by a scale (R_L, in cm). The shoot dry mass (S_M) and roots mass (R_M) were determined after drying in an oven at 70°C during 48 h, followed by weighing on a balance. The ratio of roots and shoots was then calculated ((S_M/R_M) × 100).

Statistical Analyses

They were determined significant differences between the treatments (*P*, DUNCAN test). All data were subjected to analysis of variance (ANOVA) with fixed factors using ***SPSS*** statistical software (version 8.0). DUNCAN's multiple range test was used to compare means at the 5% level. All the germination experiments were carried out in three independent experiments and the results were expressed as mean values ± standard deviation. The gas exchange and growth measurements were expressed as mean values ± standard deviation of three independent experiments.

RESULTS

Effect of Pulp, Temperature and NaCl on Seed Germination

The presence of the pulp influenced germination and number of days necessary for the germination of seeds (*G*). Scarification of *P. atlantica* seeds decreased the period to one week, as shown in Fig. 1. The manual scarification facilitated germination but the percentage does not exceed 10% for *P. atlantica* and 20% for *P. lentiscus*. On the contrary, the scarification helps the germination of harvested seeds. Statistical analyses showed highly significant differences ($P \leq 0.001$) for the effect of the pulp for both species. The germination percentage (*G*) and seed dormancy (*D*) were strongly influenced by temperature ($P \leq 0.001$). Seed dormancy is 21 days at 25°C for *P. atlantica*, and 23 and 27 days at 25 and 20°C for *P. lentiscus*, respectively. The lowest values were recorded for *P. atlantica*. At 20°C, germination is 0 and 5% for *P. atlantica* and *P. lentiscus*, respectively; but at 25°C, *G* is 10% for *P. atlantica* and 19% for *P. lentiscus* (Fig. 2). Figure 3 shows that germination is variable according to species ($P \leq 0.001$) and NaCl concentrations ($P \leq 0.001$). Salinity reduced germination for both species and increased dormancy in *P. atlantica*, which maintains lower germination than *P. lentiscus*, not exceeding 10% for all NaCl concentrations.

Responses of Seedlings to Water Deficit

There is a highly significant effect on physiological parameters of seedlings for species ($P \leq 0.001$). Both

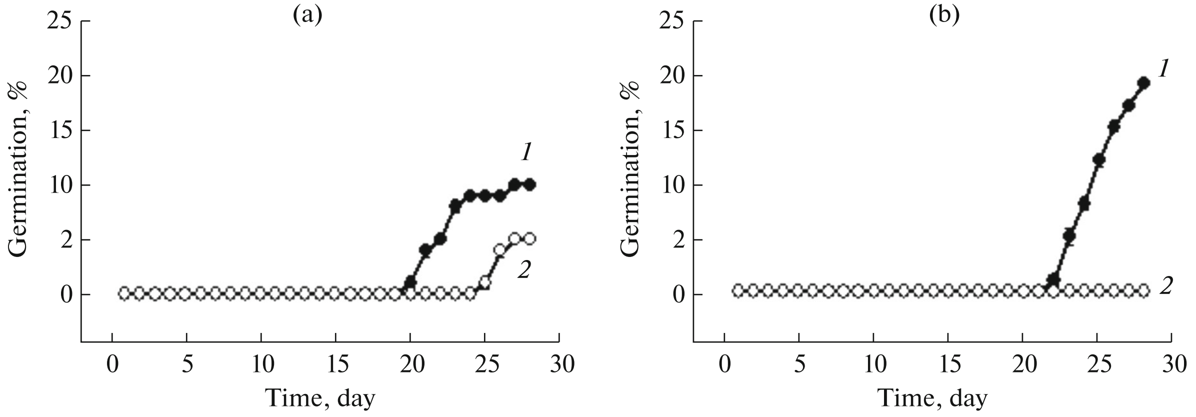

Fig. 1. Variation in cumulative percentage of germinated seeds as a influenced by the pulp for non-scarified seeds of *P. atlantica* (a) and *P. lentiscus* (b); without pulp (*1*); with pulp (*2*); $n = 6$; $P \leq 0.05$.

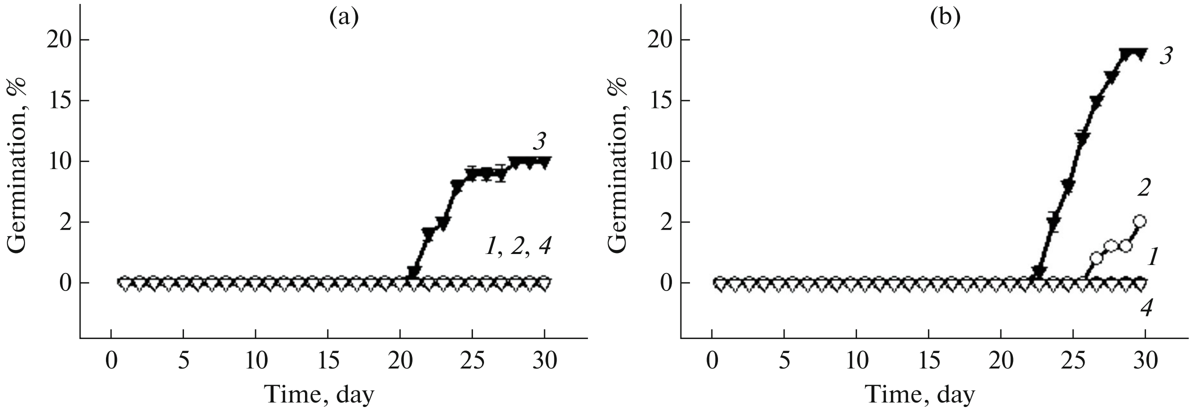

Fig. 2. Variation in cumulative percentage of germinated seeds in response to temperature for non-scarified seeds of *P. atlantica* (a) and *P. lentiscus* (b); 15 (*1*); 20 (*2*); 25 (*3*); 30 (*4*); $n = 6$, $P \leq 0.05$.

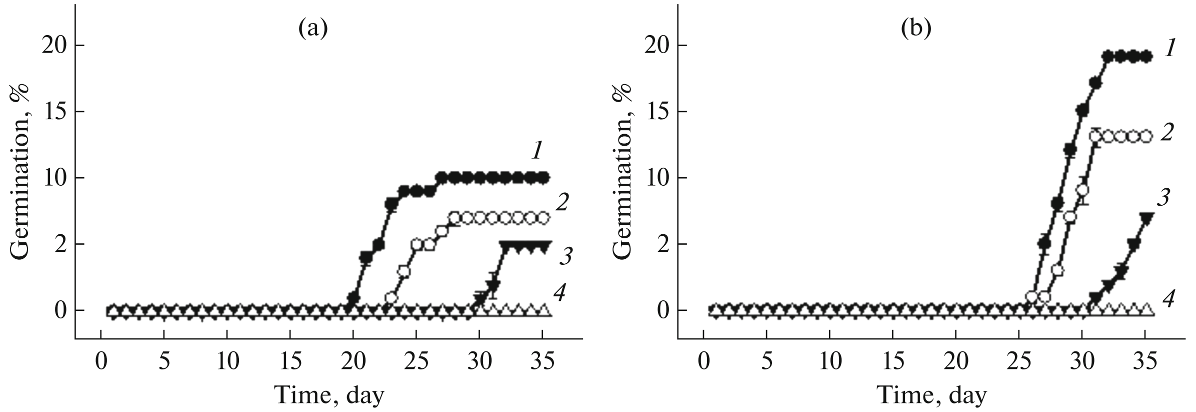

Fig. 3. Variation in cumulative percentage of germinated seeds in response to NaCl for non-scarified seeds of *P. atlantica* (a) and *P. lentiscus* (b); 0 mM (*1*); 10 mM (*2*); 25 mM (*3*); 50 mM (*4*); $n = 6$; $P \leq 0.05$.

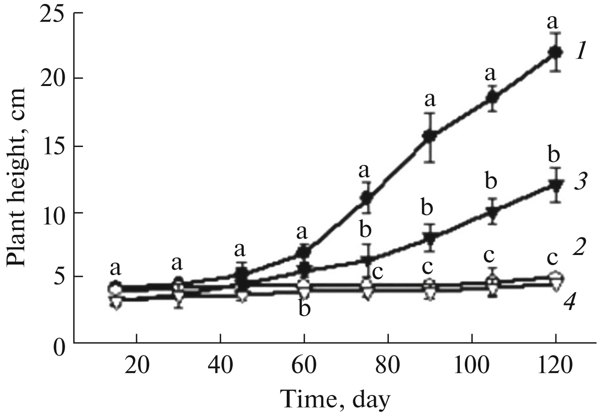

Fig. 4. Variation in plant height of seedlings of *P. atlantica* and *P. lentiscus* subjected to two water regimes: Irrigated and drought; *P. atlantica* irrigated (*1*); *P. atlantica* stressed (*2*); *P. lentiscus* irrigated (*3*); *P. lentiscus* stressed (*4*); $n = 6$; $P \leq 0.05$.

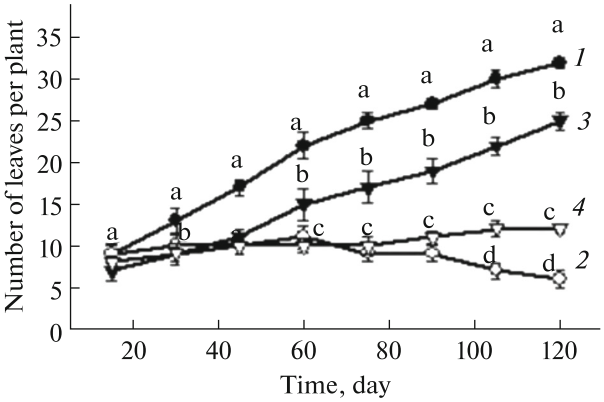

Fig. 5. Variation in the number of leaves in seedlings of *P. atlantica* and *P. lentiscus* subjected to two water regimes: Irrigated and drought; *P. atlantica* irrigated (*1*); *P. atlantica* stressed (*2*); *P. lentiscus* irrigated (*3*); *P. lentiscus* stressed (*4*); $n = 6$; $P \leq 0.05$.

species had increased stomatal conductance (g_s) under the control regime. However, water deficit greatly reduced stomatal conductance (g_s, $P \leq 0.001$), at second stage, by 89 and 69% for *P. atlantica* for *P. lentiscus*, respectively. Water deficit significantly reduced net photosynthesis rate (A) ($P \leq 0.001$) by 85 and 54% for *P. atlantica* and *P. lentiscus*, respectively. Similarly, water deficit significantly reduced the transpiration rate (E) ($P \leq 0.001$), at second stage, in the seedlings of *P. atlantica* and *P. lentiscus* (Table 1).

Water deficit had an effect ($P \leq 0.01$) on the morphological traits of *P. atlantica* and *P. lentiscus* seedlings. Under irrigated conditions, seedling height for *P. atlantica* and *P. lentiscus* reached 22 and 12 cm, respectively, at the end of the test. Water deficit reduced seedling height ($P \leq 0.001$), especially for *P. atlantica*, where the stressed seedlings did not exceed 5 cm (Fig. 4).

In control seedlings, *P. atlantica* produced more leaves than *P. lentiscus* (Fig. 5). However, under water deficit, *P. atlantica* had more reduced leaf mass and displayed more leaf senescence than *P. lentiscus*. At harvest, leaves were reduced to 6 in *P. atlantica* and 12 in *P. lentiscus* (Fig. 5). According to the data, the differences observed between species for the leaf production were significant ($P \leq 0.001$).

As a result of leaf morphological changes in *P. atlantica* and *P. lentiscus*, the leaf area was reduced by 87 and 80%, respectively, ($P \leq 0.001$) (Table 2). Under water deficit, *P. atlantica* developed deeper roots than the irrigated seedlings (69%). However, no significant effect was noted for the root length of *P. lentiscus* ($P \leq 0.001$). The stressed seedlings reduced the root mass (R_M) in stage 2 ($P \leq 0.001$), corresponding to a reduction by 84% for *P. atlantica* and 65% for

Table 1. Variation in stomatal conductance (g_s), the net photosynthesis rate (A), the rate of transpiration (E), the ratio between the net photosynthesis rate and the rate of transpiration (A/E) and the relative water content (RWC) in seedlings of *P. atlantica* and *P. lentiscus* subjected to two water regimes (irrigated and drought)

Species	Stage	Water regime	g_s, mol CO_2/m^2 s	A, μmol CO_2/m^2 s	E, mmol H_2O/m^2 s	A/E, μmol CO_2/mmol H_2O	RWC, %
P. atlantica	Stage 1	Well irrigated	0.15 ± 0.02^{ab}	12.70 ± 1.09^{ab}	6.33 ± 0.34^{a}	2.06 ± 0.75^{b}	87.12 ± 6.34^{a}
		Drought	0.13 ± 0.01^{bc}	10.70 ± 1.11^{bcd}	4.28 ± 0.34^{d}	2.50 ± 0.81^{ab}	84.08 ± 5.17^{a}
	Stage 2	Well irrigated	0.18 ± 0.01^{a}	13.64 ± 0.59^{a}	5.23 ± 0.6^{bc}	2.60 ± 0.40^{ab}	82.87 ± 7.53^{a}
		Drought	0.02 ± 0.01^{e}	2.30 ± 0.57^{f}	1.60 ± 0.2^{e}	1.44 ± 0.42^{b}	52.67 ± 6.10^{b}
P. lentiscus	Stage 1	Well irrigated	0.09 ± 0.02^{d}	9.90 ± 0.62^{cd}	5.60 ± 0.50^{ab}	1.67 ± 0.65^{b}	83.45 ± 4.03^{a}
		Drought	0.10 ± 0.02^{cd}	9.38 ± 0.48^{d}	5.70 ± 0.30^{ab}	1.54 ± 0.39^{b}	86.82 ± 7.59^{a}
	Stage 2	Well irrigated	0.13 ± 0.01^{bc}	11.18 ± 0.95^{bc}	4.47 ± 0.39^{cd}	2.50 ± 0.97^{ab}	80.03 ± 6.41^{a}
		Drought	0.04 ± 0.01^{e}	5.12 ± 0.20^{e}	1.50 ± 0.36^{e}	3.41 ± 0.58^{a}	57.89 ± 4.60^{b}

Different superscript letters indicate statistically significant parameter changes during the year at $P \leq 0.05$ (analysis of variance ANOVA, Duncan's test, $n = 6$).

Table 2. Variation in leaf area (L_A), root length (R_L), root mass (R_M), the shoot dry mass (S_M), the ratio between shoot dry mass and root mass (S_M/R_M) in seedlings of *P. atlantica* and *P. lentiscus* subjected to two water regimes: (irrigated and drought)

Species	Stage	Water regime	L_A, cm^2	R_L, cm	R_M, g	S_M, g	S_M/R_M, %
P. atlantica	Stage 1	Well irrigated	15.40 ± 1.36^c	19.60 ± 2.57^c	0.13 ± 0.01^e	0.14 ± 0.01^{de}	1.12 ± 0.06^b
		Drought	16.38 ± 1.06^c	20.80 ± 1.80^c	0.14 ± 0.02^e	0.16 ± 0.02^{cd}	0.87 ± 0.05^c
	Stage 2	Well irrigated	90.35 ± 5.73^a	54.00 ± 2.70^b	0.52 ± 0.01^a	0.77 ± 0.02^a	0.65 ± 0.09^d
		Drought	11.70 ± 0.40^d	91.40 ± 4.95^a	0.48 ± 0.01^b	0.12 ± 0.01^e	4.51 ± 0.06^a
P. lentiscus	Stage 1	Well irrigated	16.61 ± 0.53^b	12.16 ± 1.30^d	0.06 ± 0.01^f	0.16 ± 0.01^{cd}	0.40 ± 0.05^e
		Drought	17.22 ± 1.53^b	13.08 ± 1.36^d	0.08 ± 0.01^f	0.17 ± 0.01^c	0.47 ± 0.06^e
	Stage 2	Well irrigated	68.40 ± 4.73^b	54.40 ± 2.57^b	0.24 ± 0.02^c	0.51 ± 0.02^b	0.45 ± 0.02^e
		Drought	13.53 ± 1.73^d	53.60 ± 2.24^b	0.16 ± 0.01^d	0.18 ± 0.01^c	1.06 ± 0.07^b

Different superscript letters indicate statistically significant parameter changes during the year at $P \leq 0.05$ (analysis of variance ANOVA, Duncan's test, $n = 6$).

P. lentiscus. The water deficit reduced the shoot dry mass (S_M) for *P. lentiscus* (33%), with a high S_M/R_M in *P. atlantica* ($P \leq 0.01$) indicative of a preferential allocation of nutrients in favor of root development.

DISCUSSION

The germination rate of the genus *Pistacia* seeds largely depends on the species. The best results were observed in seeds without pulp for *P. atlantica* [1], which underwent mechanical pulping [27]. The pulp is an impermeable barrier that must be scarified mechanically or chemically to facilitate passage of water necessary for germination [12]. In this context, seed germination in *Ceratonia siliqua* and *P. lentiscus* [27], and *P. vera*, was improved by scarification pretreatment [28]. In the present study, the pulp prevents germination in *P. lentiscus* and increases the period of germination in *P. atlantica*. Matured fruits of *P. atlantica* have attractive colors for various frugivores that ensure their dispersal and pulping of seeds [5]. Furthermore, the pretreatment of seed carried by birds has an effect on seed dormancy and seed germination percentage [29]. This may be explained by the fact that seed dispersal and germination are closely linked to specific frugivores. Seed germination is regulated by genotypic and environmental characteristics such as temperature. Germination tests performed here for *P. atlantica* and *P. lentiscus* have shown that germination percentage and the rest period are highly related to temperature [11]. The best rate of performance is obtained at 25°C. According to *Bareke* et al. [12] *and* Luna et al. [30], temperature can affect germination by seed deterioration, loss of dormancy and by the germination process itself. Both species are sensitive to high salinity. In fact, according to Ungar et al. [31], the inhibition of germination by NaCl is not due to the toxic effect of Na^+ ions, but rather to the osmotic effect of NaCl, which prevents imbibition or inhibits hormonal or enzymatic activity. Therefore, *P. lentiscus* accessions are potential promising material for the saline soil valorization [32]. In this study, the response of *P. atlantica* and *P. lentiscus* seedlings to water deficit resulted in physiological and morphological changes that affect plant water status, photosynthetic gas exchanges, resource allocation and growth. Relative water content (RWC) was used to evaluate plant water status [33]. The success of seedling establishment and growth depends on stored soil moisture to ensure survival during the drought period. Thus, plants must establish root systems at depths where water is available [14]. To the best of our knowledge, research on the relationship between root morphology and the ability to transport water in Mediterranean woody species in response to drought is still scarce [16, 18]. The study of leaf gas exchange has revealed that under water stress, photosynthesis and transpiration are substantially reduced in *P. atlantica* and *P. lentiscus* seedlings following stomatal closure [16, 34]. Moreover, during severe or prolonged stress, inhibition of the mesophyll activity can be linked to lower stomatal conductance (g_s) in Mediterranean sclerophytes [15]. The more stress is accentuated, the more the metabolic regulation of photosynthesis increases, and the assimilation of CO_2 decreases continuously as a function of the severity of the applied water stress [9]. Stomatal control is reported in stressed seedlings as a response to a message from hormonal action in roots, thus limiting the availability of CO_2 in the substomatal chamber [8, 35]. Based on the results of our study, we conclude that the decrease in stomatal opening and limitations on photosynthesis, followed by the decrease in individual leaf area, are the main causes of the reductions in carbon uptake and whole biomass seedlings, based on reasoning presented in a previous report [17]. This response is accompanied by a reduction of leaf transpiration as an avoidance action against cell dehydration, which agrees with the results obtained in olive trees [20, 34, 36]. In *P. lentiscus*, stomatal and non-stomatal controls affect leaf gas exchanges, leading to improvement of instantaneous water use efficiency [9, 17, 37], but damages in the mesophyll activity affect photosynthesis rather than transpiration for *P. atlantica*, limiting the instantaneous water use efficiency [18]. Specific correlation between leaf water status and growth was observed. Drought effects might also have been mitigated by reduction in leaf area via leaf abscission or via limiting new needle expansion. Under water deficit, reduction in shoot growth is accompanied by reductions in the size and number of leaves, as a result of limited cell division and elongation in response to water scarcity [19]. The decrease in stomatal conductance and plant leaf area, either by the production of new small leaves for *P. lentiscus* evergreen species, or by progressive leaf senescence for *P. atlantica* deciduous species, is an avoidance mechanism against dehydration. In *P. atlantica*, the senescence of leaves acts to improve plant water status and the recycling of resources in the plants [9, 38]. At the establishment stage, *P. atlantica* acts by increasing stomatal resistance, reducing the absorption of CO_2, and limiting leaf and shoot growth [8]. The radical change in the allocation of resources to support more root development reflects a highly adaptive response to drought [19, 39]. In this context, the changes in biomass partitioning and balance in shoot transpiration area versus root absorption area influence subsequent seedling growth and survival [14, 16]. The stressed *P. atlantica* seedlings developed longer roots than *P. lentiscus*, which allows the reduction of the effect of water deficit in marginal and nutrient-poor soils [18, 23]. This is because the rapid development of roots improves water uptake and nutrient transport, and facilitates seedling survival and growth [18, 24]. The stage of germination of *P. atlantica* and *P. lentiscus* is critical, and germination conditions largely determine the percentage of germinated seeds [1, 12]. The presence of the pulp acts as a physical barrier which limits seed imbibition and reduces gas exchange. For this reason, mechanical or chemical pulping is advisable in order to achieve good germination for both species [1]. Indeed, our results show that, for good germination, the growth medium should not have salinity levels exceeding 50 mM NaCl and temperature must be between 20 and 25°C for seeds of *P. lentiscus*, and 25°C for seeds of *P. atlantica* [30]. It will remain increasingly important to identify the limitations to the seedling establishment in *P. atlantica* and *P. lentiscus* and act to overcome them. The *P. atlantica* seedling stage is sensitive to soil water deficit [14] and the present study showed a high-water requirement in this species. Stomatal control seen here for *P. lentiscus* and *P. atlantica* has been reported as a mechanism of dehydration avoidance in several Mediterranean species, related to the decrease of water loss through the stomata [8, 15, 18]. Plants with longer roots have the ability to tolerate drought, with root morphology being an effective indicator of seedling performance [5, 14, 16]. For this reason, in reforestation programs, poor performance of planted seedlings is also associated with poor soils and soil water deficit [14, 39]. In contrast, our study shows that *P. lentiscus* seedlings had values closer to control values, reflecting its tolerance to water deficit. The success of *P. lentiscus* seedlings is attributed to the maintenance of higher photosynthetic activity and leaf growth [21, 24]. In this context, the survival of *Pistacia lentiscus* and *Pinus halepensis* seedlings was positively correlated to initial seedling root collar diameter and the total dry weight [24]. Besides, many other morphological characteristics were correlated with field success, such as seedling diameter and shoot height [14, 39]. The restoration of Mediterranean and degraded areas is often unsuccessful and this is partly due to insufficient knowledge of the ecology of native species at the seedling stage [25]. Under natural conditions, the distribution of *P. atlantica* and *P. lentiscus* may be limited by low seed germination, drought and skeletal soils poor in nutrients [8]**.** The divergence within *P. lentiscus* and *P. atlantica* seedlings in seed germination, carbon assimilation, growth and biomass allocation are interpreted as a plasticity response, indicating a specialization to either original or resource habitats [8, 23].

ACKNOWLEDGMENTS

We thank the technical staff of the Laboratory of Plant Physiology at the Faculty of Sciences of Gafsa (Tunisia) who carried out the experiments on the seeds.

FUNDING

This study was financially supported by the bilateral Tunisian-French project PHC-(12/G0910) managed by the CMCU.

COMPLIANCE WITH ETHICAL STANDARDS

The authors declare that they have no conflicts of interest. This article does not contain any studies involving animals performed by any of the authors.

CONFLICT OF INTEREST

The authors declare that they have no conflicts of interest.

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