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Effects of seed sources and shade on vigor of Brant's oak seedling

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The use of local seed provenance is often recommended in forest restoration. Early vigor is a combination of the performance of seed germination and emergence after planting. The ability of young Brant's oak plants to grow and develop after emergence and its dependence on local habitat conditions was investigated in this study. The effects of seed source and shade on early growing seedlings of Brant's oak (*Quercus brantii* L.) were determined in field measurements. Seeds of *Quercus brantii* L. were collected from 4 forest areas (seed sources) in southern Zagros (Provinces of Kohkilouyeh-Bouyer Ahmad and Fars) at altitudes of 850, 1,100, 1,500, 2,100 m a.s.l., and planted in a nursery constructed in southwestern Iran. According to a split-plot design consisting of four blocks, each containing two main treatment plots (no shading, partial shading), each main plot was sub-divided into four sub-plots (for elevations of 850, 1,100, 1,500 and 2,100 m). Results showed that shade treatments had significant effects on emergence percentage and rate, shoot length, shoot dry weight (SDW), root dry weight (RDW), leaf area (LA), and chlorophyll content. Ecological factors also had an effect on seed performance. Altitude of seed source had a very significant effect on root length, LA, SDW, and RDW. The seeds collected from 850 m a.s.l. elevation showed the highest performance, especially in leaf area, root length, shoot dry weight, and root dry weight. Our results showed that the altitude of 850 m a.s.l. was the best for collecting Brant's oak seeds.

Key words: altitude, Brant's oak (Quercus brantii L.), emergence percentage, Iran, root, seed source, shading, shoot

INTRODUCTION

Approximately 60% of Iran is classified as arid and semi-arid. Much of the country has a desert climate, with an average annual precipitation of less than 300 mm (Mansoory 1992). The zagros forests of Iran are classified as semi-Mediterranean forests because of their geographic location and the effects of Mediterranean climate on it. Zagros mountains are the original source of *Quercus libani* and *Q. boissieri*. Brant's oak (*Quercus brantii* L.) is one of the most important deciduous tree species of dry forests in Iran (Zohary 1973). The annual precipitation of the zagros range of the Fars-Zagrosian Brant's oak forest varies from about 400-800 mm, falling mostly in the winter and spring (McGinley 2008). The climate

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is harsh, with very cold winters and hot dry summers. Four or more months of the year have mean minimum temperatures below 0°C. Human activities and domestic animals have affected dry forests, severely limiting tree growth, and reducing the extent of these forests by cutting and grazing (El-Moslimany 1986).

Brant's oak only regenerates by seed. Seed quality is also critical for the early vigor of a new plant. Early vigor is a combination of the ability of the seed to germinate and emerge after planting, and the ability of the young plant to grow and develop after emergence (Jin and Wang 2002). Seed and uniformity of germination are limiting factors in forest trees (Fenner 1992). Seed germina-

Received 22 April 2010, Accepted 01 October 2010 *Corresponding Author

E-mail: taghvaei@shirazu.ac.ir Tel: +98-711-613-8163 tion plays an important role in the regeneration of plant species, especially under unpredictable environmental conditions like those in Mediterranean ecosystems (Giménez-Benavides et al. 2005). Poor germination and seedling establishment are regarded as the major causes of low densities in Mediterranean forests (Close and Wilson 2002). Seed performance also depends on vigor (Copeland and McDonald 1995). Seed vigor can be affected by provenance or by environmental factors operating during development and maturation, such as seed germination, seedling establishment, and survival regulated by light, climate, edaphicity, and species-specific factors (Taghvaei 2006, Liu et al. 2008).

Environmental factors such as temperature affect germinability of seeds of many species during seed maturation (Pourrat and Jacques 1975). In different plant species, even small differences in temperature during plant development or seed maturation can influence the germinability of seeds (Gutterman 1996). Light quality affect on germinability during seed filling too. Altitude can also influence germinability of seeds; the higher the altitude of seeds, the lower their rate of germination is (Dorne 1981). Seed provenance affects the seed germination, seedling survival, and growth of Prosopis cineraria L. (Arya et al. 1995), the growth and flowering of Coreopsis lanceolata and Salvia lyrata (Liu et al. 2008), and the seed germination, vigour index, germination rate index and oil content in sunflowers (Norcini et al. 2001). Thus, any study of forest restoration should ideally separate the effects of individual environmental factors from their interactions with demographic stages. Early growth stage is an important stage, affected by environmental factors during seed germination and emergence. Germination percentage, time-to-germination, seedling survival, height, flushing, and number of secondary branches of Abies guatemalensis (Rehder), show significant differences among provenances (Andersen et al. 2008).

In tropical dry forests, germination, survival, and seedling growth are affected by light intensity (Khurana and Singh 2001). The reflex of germination to light intensity depends on the species and environment. Biomass production, relative growth rate, root/shoot ratio, specific leaf area (LA), net assimilation rate, and LA ratio can be limited by either poor light availability (Rincón and Huante 1993) or low soil moisture due to water use by nurse trees (Rodríguez-Calcerrada et al. 2008) in the dry forest understory. The shade protects soil moisture during seed germination in dry forests. Also it has been found that germination success was approximately equal in shaded and un-shaded sites for some species (Ray and Brown 1995). In dry forests, species germination and early establishment must occur during the first of the wet season when water is at its most available (Khurana and Singh 2001). The general assumption of this study is that provenance variables are a complex of environmental factors and shade changes in early stages of the life cycle of an acorn seed. The aim of this study was to test whether seed vigor and seedling performance are affected by altitude seed source and shade during seedling establishment.

MATERIALS AND METHODS

Plant material

Seed lots of Brant's oak (*Quercus brantii* Lindl.) were collected from 4 forest habitats (seed sources) in southern Zagros (Provinces of Kohkilouyeh-Bouyer Ahmad and Fars), located at elevations of 850, 1,100, 1,500, and 2,100 m a.s.l. in Iran. After collection, acorns were stored within bags in a refrigerator (5°C) until the start of each experiment.

Experimental design

The experiment was set up within an intact area of the College of Agriculture (Badjgah 29°50' N, 52°46' E) at Shiraz University, located at an elevation of 1,810 m a.s.l. in the southwest of Iran (Table 1). The experimental de-

Table 1. Average of maximum, minimum of temperature and monthly of total of precipitation of Badjgah

	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec
Maximum temperature (ºC)	12.84	12.81	16.47	23.91	27.83	34.09	35.61	35.08	32.10	28.58	19.14	13.58
Minimum temperature (ºC)	-5.79	-2.67	1.49	4	6.75	10.75	14.52	14.36	10.47	5.05	3.21	-3.36
Precipitation (mm)	0.68	1.05	0.77	0.11	0.00	0.00	0.00	0.00	0.00	0.00	1.40	0.42

sign was a split plot one based on a complete randomized block with four blocks: two light levels (no shading, partial shading) were assigned to the main plots, while four seed source sites (altitudes of 850, 1,100, 1,500, 2,100 m a.s.l.) were assigned to the sub plots. There were two types of shade: no shade (full sunlight) and 50% shade by shade cloth. The shade used after seed planting to final experiment.

To determine the effects of provenance and light on the germination of seeds and the early establishment of seedlings, the seeds from each treatment were planted in a 0.5-L plastic pot filled with 500 g of sandy loam soil. The pots were watered weekly with sufficient water. Emerging seedlings were counted every three days, continuing until when no further germination was recorded, and then vigor-related traits were measured on individual plants after five months. The traits were: emergence percentage (EP), shoot length (SL), and root length (RL). Chlorophyll content (CC) was recorded using the lowest seedling leaf fully expanded from the top of the main stem for each pot at 10:00-12:00 a.m. by a SPAD chlorophyll meter (Minolta SPAD-520; Minotal, Ramsey, NJ, USA) by LA (cm²) at the end of this period. LA was measured on collected leaves of the shoot with a LA meter, model MK2 (Delta-T Co., Cambridge, UK). SL was measured by the length from the top to the shoot, and RL was measured by the length from the top of the root. Shoot dry weight (SDW) and root dry weight (RDW) were measured after drying for 24 h in an oven at 70°C (ISTA 1999). Mean time of emergence (MTE) and emergence rate (ER) were calculated for each treatment according to the equations of Ellis and Roberts (1981) and Agrawal and Dadlani (1992) as follows, respectively:

> $MTE = \sum (ni.ti) / \sum n$ $\text{ER} = \sum (ni/ti)$

Tal	ble	2.	Analysis o	fvariance	for seed	and	seedling	parameters
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MTE: mean time to full emergence ER: emergence rate

Where ni is the number of emerged seeds per day, ti is the amount of time counted from the beginning of emergence. CC was measured on the lowest seedling leaves with a Minolta chlorophyll meter.

Data analyses

Data were checked for normality, and then were analyzed using MSTATC statistical software (MStat Inc., East Lansing, MI, USA). Treatment means were separated by a Duncan test if the F-value of the treatment was significant at the 0.05 or 0.01 probability levels.

RESULT

Emergence percentage (EP)

EPs were significantly different between light treatments (P < 0.05) but not between seed sources or sites (Table 2). The mean EPs were 88% and 65% in light and shade respectively (Fig. 1). In treatments of all altitudes, shade treatment decreased the EP. The highest seed EP (96%) was obtained at an altitude of 850 m above sea level. Seed emergence decreased as the altitude increased (Fig. 1).

Emergence rate (ER)

There were significant differences in ERs between light treatments (P < 0.01) (Table 2). The mean ERs were 4.61 and 1.86 in light and shade, respectively (Fig. 2). There was no significant difference in ER between seed sources

Table 2. Analysis of variance for seed and seeding parameters											
Source	Degrees of freedom	EP	ER	MTE	SL	RL	RDW	SDW	LA	CC	
Replication	1	153.78	3.88	1.88	4.39	3.44	0.029	0.021	115.17	5.72	
Shade (A)	3	2317.61^{*}	50.73**	7.84	433.72**	27.67	0.69^{*}	0.25^{*}	351.85^{*}	250.26^{*}	
Error	3	116.36	1.24	1.32	3.04	11	0.06	0.007	25.83	16.08	
Seed source (B)	3	387.20^{*}	1.18^{*}	6.10	10.21	23.22^{*}	0.41^{*}	0.21^{*}	648.17	23.48	
A [*] B	3	968.67	3.27	8.66	0.983	1.37	0.053	0.01	57.95^{*}	12.57	
Error	18	228.59	0.80	4.71	8.05	1.731	0.05	0.015	143.74	12.25	

^{*} and ^{**} indicate significance of P < 0.05 an P < 0.01, respectively.

EP, emergence percentage; ER, emergence rate (per day); MTE, mean time emergence; SL, shoot length (cm); RL, root length (cm); RDW, root dry weight (g); SDW, shoot dry weight (g); LA, leaf area (cm²); CC, chlorophyll content (spad).



 $Fig. \ 1.$ Emergence percentage of $\textit{Quercus brantii}\ L$ as affected by seed source and light density.



Fig. 2. Emergence rate of *Quercus brantii* L. as affected by seed source and light density.



Fig. 3. Mean time emergence rate of Quercus brantii L. as affected by seed source and light density.



 $Fig.\ 4.$ Shoot length of $\textit{Quercus brantii}\ L$ as affected by seed source and light density.



 $Fig. \ 5.$ Root length of $\it Quercus \ brantii$ L. as affected by seed source and light density.



 $Fig. \ 6.$ Shoot dry weight of $\it Quercus \ brantii$ L. as affected by seed source and light density.



Fig. 7. Root dry weight of $Quercus \ brantii$ L. as affected by seed source and light density.



Fig. 8. Leaf area of *Quercus brantii* L. as affected by seed source and light density.



Fig. 9. Chlorophyll content of *Quercus brantii* L. as affected by seed source and light density.

or sites, but the highest seed ER (5.32) was obtained for altitudes of 850 and 1,100 m above sea level (Fig. 2).

Mean time of emergence (MTE)

There were significant differences in MTE between light treatments (P < 0.05) (Table 2). The shade treatment had a longer MTE than light. The MTE were 4.9 and 5.78 days in light and shade, respectively (Fig. 3). There was no significant difference in MTE between seed sources or sites, but the lowest MTE (4.7 days) was obtained at an altitude of 850 m above sea level (Fig. 3).

Shoot length (SL)

SLs were significantly affected by light treatment (P > 0.01) (Table 2) but not by seed sources or sites. Shade treatment increased SL in all treatments. SLs were 12.71 cm and 20.08 cm in light and shade, respectively (Fig. 4).

Root length (RL)

There were significant differences in RLs between site treatments (P < 0.01), but not between light treatments (Table 1). However, the light treatments had a longer RL than shade (Fig. 5). The longest RL was produced by seeds that were collected at altitudes of 850 and 1,100 m above sea level (Fig. 5).

Shoot and root dry weights (SRDWs)

SRDWs were significantly affected by light (P > 0.05) as well as seed sources and sites (P > 0.01) (Table 2). Shade treatment increased SDW significantly in all treatments (P > 0.05). SDWs were 0.91 g and 0.73 g in light and shade, respectively (Table 2). RDW in light was significantly higher than shade treatment (Fig. 6). RDWs were 1.07 g and 0.81 g in light and shade, respectively (Fig. 6). The highest root and shoot weights were produced by seed that collected at an altitude of 850 m above sea level (Figs. 6 and 7).

Leaf area (LA)

There were significant differences in leaf areas between light treatments (P < 0.05) (Table 2). The leaf areas were 58.5 and 60.86 cm² in light and shade respectively (Fig. 8). There was no significant difference in LA between seed sources or sites. But the highest LA (72.07 cm²) was obtained in the altitude of 850 m above sea level (Fig. 8).

Chlorophyll content (CC)

CC differed significantly between light treatments (P < 0.05), but not between seed sources or sites (Table 2). CCs were 43.46 SPAD and 49.04 SPAD in light and shade, respectively (Fig. 9). Shade treatment increased the CC for treatments at all altitudes (Fig. 9).

DISCUSSION

Seed source

Source variation tests is one of method to screen the naturally available genetic variation to select the best planting material for higher productivity (Bhat and Chauhan 2002). The seed may respond to changes in environment (Egli 1998). This study has revealed that the present provenance is a factor controlling the performance of Quercus brantii L. Seeds. The highest seed EP and ER were obtained at an altitude of 850 m above sea level, and both of them decreased as the altitude increased. Shivanna et al. (2007) reported significant differences among seed sources in seed germination and seeding traits. Latitudinal differentiation on growth characteristics has also been observed in a number of studies on conifers (Wright 1976, Hansen et al. 2004). Therefore, alternation of provenance altitude was found to be an effective factor on seedling parameters. Increase of altitude above sea level decrease seed vigor (EP and ER) and seedling performance (LA, CC, RL, SL, SRDW, etc.). Our results detected that seedling growth decreases with increasing altitude, and that higher altitudes have lower emergence, ER, RL, SL, SRDW, and LA. A similar trend of decreasing seedling growth with increasing altitude has been detected in provenance tests of Abies species (Bongarten and Hanover 1986), but Krishan and Toky (1996) reported that there was no significant relationship between Acacia nilotica ssp. indica seed germination and seedling growth with the latitude or longitude of the original seed source. Jin and Wang (2002) also reported that Bidense pilosa L. var radiate seeds at high altitude showed higher germination percentage, speed of germination, and shorter mean days of germination. Growth season and seed filling duration at low altitudes is longer than at high altitudes; therefore, allowing complete maturation and material translation to seed are necessary for seed vigor (Egli 1998). Early seed vigor is an important trait in forest improvement. Early vigor is a complex of plant performance that is reflected in variations in plant

characteristics such as EP, ER, LA, CC, RL, SL, SRDW, etc. (Norcini et al. 2001, Andersen et al. 2008). Seed emergence and seedling performance strategies have important roles in the regeneration of plant species, especially under theunpredictable conditions of Mediterranean ecosystems. So, for dry forest conservation, low altitudes are the best locations for seed harvesting.

Shade treatment

Forest production differs from in open areas and under forest canopy (Chen et al. 1995). In this experiment, shade treatment had an influence on acorn germination, seedling emergence, and seedling growth. EP, ER, SL, RL, LA, and CC field were significantly affected by light treatments . In general, shade makes for a micro site that may be suitable or not suitable for acorn germination and seedling emergence. Acorns in such sites were protected from humidity and temperature fluctuations (Sarlöv-Herlin and Fry 2000). Shade treatment increased SL, RL, shoot weight, LA, and CC. Factors influencing early survival and growth of seedlings underground may be very different from those determining the growth of older seedlings. Early growth of Brant's oak seedlings under ground in shade or light depend primarily on stored food reserves in the acorns and not on current materials of photosynthetic production (Crow 1988). Once cotyledon reserves are depleted, the growth and survival of seedlings depend on photosynthetic produce generated by new leaves. Light quality then becomes a limiting factor for survival and growth (Crow 1988). Although light is a dominant environmental factor limiting seedling establishment, shade treatment may increase root and shoot growth. Root regeneration and root growth are very sensitive to soil moisture stress (López-Barrera and Newton 2005). In northern red oak, root initiation and growth has been shown to cease at soil osmotic potentials between -0.4 and -0.6 MPa (López-Barrera and Newton 2005). Shade treatment decreased EP and ER. The shade protects soil moisture and decreases soil temperature, so it decreases the field emergence of seeds (Charlton et al. 1986). Additionally, germinated and non-germinated acorns sowed in shade treatments are less damaged by insects compared to light treatments (López-Barrera and Newton 2005). Ray and Brown (1995), however, found that germination success in dry forests in the US Virgin Islands was approximately equal in shaded and unshaded sites. In dry forest, species germination and early establishment must occur during the wet season when water is more available. Therefore, the canopy modifies

the light quality at the ground level by its filtering influence (Rincón and Huante 1993), and thereby improves survival after emergence in dry seasons (Lieberman and Li 1992).

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