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Growth variability in *Argania spinosa* seedlings subjected to different levels of drought stress

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Argan tree, the only representative species of the tropical family Sapotaceae in Morocco is distributed in arid and semi-arid areas. Tolerance to drought remains poorly described for this species; we applied five levels of drought stress and monitored growth variables and biomass production of seedlings in pot and in field propagated from seeds of eight genotypes from three geographical origins. Drought stress decreased seedlings height, basal diameter, leaf number, leaf area, and biomass production and water content but increased root / shoot length. Even under moderate or severe drought stress (1/4 and 0 FC), some genotypes (mother-trees) sustained shoot growth, they have taproots exceeding 50 cm, which are accompanied by a large number of lateral roots, more leaves and great leaf areas. But others showed an increase below-ground length and biomass indicating a higher root / shoot ratio under drought stress conditions. So, in order to enhance the survival rate and conserving rate of seedlings planting, appropriate human intervention is required to reduce the damage to Argan seedlings resulting from drought.

Key words: *Argania spinosa*, water stress, aridity, shoot, root, seedling biomass production.

INTRODUCTION

Argan tree, the only representative species of the tropical family Sapotaceae in Morocco is distributed in arid and semi-arid areas. It plays major roles against soil erosion, desertification and preservation of biodiversity and in the daily life of local populations (Emberger, 1924; M'Hirit et al., 1998; Msanda et al., 2005). Currently, it is proven that the adult tree is drought resistant, survives in its area for many years (Emberger, 1925, 1955; Boudy, 1952) and produces branches, leaves, flowers and fruit with a rainfall exceeding 100 mm (Ferradous et al., 1996; Zahidi, 1997a, b; Bani-Aameur, 2001). In this ecological environment quite fragile, degradation of Argan is a dynamic process progressive owing to increasing population, high demand for agricultural and overgrazing. Therefore, conservation and regeneration of this tree are a regional priority for protection against the increased desertification (Bani-Aameur, 2007). Argan tree is regenerated by seed and release. Regeneration by seedlings allows preservation of genetic diversity of this species. However, fruit harvesting, livestock grazing of few seedlings issued from germination of some remaining kernels (Boudy, 1950, 1952), action of aridity on seed germination and difficult conditions of seedlings survival after germination are factors limiting natural regeneration from seed (Zahidi and Bani-Aameur, 1998; Bani Aameur and Alouani, 1998).

Artificial regeneration is the only possibility that can ensure the survival and maintain an appropriate level of genetic diversity of the species. If germination has been very successful, since percentage of germinated seeds was more than 80% (Bani-Aameur and Alouani, 1999), testing in field of transplanted seedlings grown in nursery

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was always doomed to failure. Traditionally, observed reforestation failures can be explained by the difficulty of seedling establishment after transplantation. Some techniques to improve transplantation rate have been tested on a small scale (Harrouni et al., 1995). They sometimes lead to improved success rates; however, they will remain below an acceptable level. In arid or semi-arid areas, during periods of drought, most plants are likely to be under stress, but differences in the degree of stress can have important implications for plant survival. One of the most general types of stress experienced by plants is water limitation. As a result of water limitation, plants express various responses and have developed a wide diversity of drought tolerance mechanisms from both morphological and physiological aspects (Blum, 1983; Loss and Siddique, 1994; Li et al., 2007). In field, the very definition of drought accepted by many authors includes a reduction in plant growth due to decrease in average precipitation amounts (Tucker and Goward, 1987). Water shortage can cause plants to reduce their metabolic activity, causing a decrease in photosynthesis, carbon fixation and ultimately growth (Younis et al., 1993; Li et al., 2007). Some studies have shown that drought stress can affect the growth of plant organs differently, which may result in the alteration of the morphological features of the plants (French and Turner, 1991).

The change in root to shoot dry mass ratio has been considered as one of the mechanisms involved in the adaptation of plants to drought stress. In order to diminish consumption and increase absorption of water, plants in dry conditions often decrease their growth rate and biomass production, and contribute more biomass to roots, so that they could maintain a higher root to shoot ratio (Yin et al., 2005; Villagra and Cavagnaro, 2006). Drought stress reduces both root and shoots growth. However, root growth seems to be less affected. Drought stress often leads to a decrease in leaf dry mass ratio (leaf dry mass/plant total dry mass) in many species (Turner, 1997). Several studies have concerned the adult tree and reported that Argan is xerophytic and can support low and irregular rainfall (Boudy, 1952; Ferradous et al., 1996). After a prolonged drought biomass production stops and the tree loses completely leaves, flowers and fruits (Bani-Aameur, 1997; Zahidi, 1997). Stomatal regulation by closure of stomata contributes very little to avoid drought (El Aboudi et al., 1991). Very few studies have focused on seedlings, according to Zahidi and Bani-Aameur (1997a, b) root length can reach 27 cm before the cotyledons opening. In seedlings grown in nursery conditions for one year, root length varied between 138.4 and 10.7 cm, stem height between 69.3 and 6.5 cm. Seedlings grown for 16.5 months were longer four times than those grown for 4.5 months, but nine times greater than those grown only for 3 months. After six months, seedlings irrigated daily at field capacity showed a gain of 9.7 cm in stem height (Harrouni et al., 1995). The decrease in water regime leads to a reduction in seedling leaf number.

In addition, in non-irrigated seedlings, root was twice longer than those of irrigated seedlings after nine months (Kaabouss, 1992; Harrouni et al., 1995). However, to ensure extension and conserving genetic diversity, especially in areas where drought can be a very important environmental factor which limit the increase of survival rate of transplanted seedlings; evaluating seedlings responses to various degrees of water stress is the key to speed up forest restoration of Argan. No specific information is available on the tolerance of seedlings to drought or on variability of growth in the difficult conditions of their natural environment.

Our objective is to test morphological responses of Argan seedlings issued from eight selected genotypes grown for 14 months in nursery and in field subjected to five water regimes.

MATERIALS AND METHODS

Plant material and growth conditions

Seeds of eight selected trees of *Argania spinosa* were collected from three geographical origins; that is, Ait Melloul Argana and Ait Baha with ecological characterization (Bani-Aameur and Zahidi, 2005; Zahidi et al., 2013). Before germination, fruit were kept in cold for one month, and then scarified as described by Bani-Aameur and Alouani (1999) before sowing (March 20 first season) in vats containing sand in a mini-greenhouse. After emergence, seedlings were transplanted (May 20 first season) in pots containing 1/3 of peat, 1/3 of sand and 1/3 of Argan loam and placed in open air at the Faculty of Sciences, Agadir. Transplantation was done with five seedlings per pot reduced to two seedlings per pot after two months of seedling growth.

In early summer (June 6th first season), seedlings were irrigated daily by five water regimes [1 field capacity (1 FC = 200 ml), 3/4 FC (150 ml), 1/2 FC (100 ml), 1/4 FC (50 ml) and 0 FC (0 ml)].

Measurements and calculations

At the end of each month, seedling height or stem length (LT cm) and basal diameter (DT cm) were recorded. The seedlings were harvested at the end of the experiment (June 6th second season), the following characters were measured: LT: Seedling height (cm), NF: leaf number on the main stem, NE: spines number on the main stem, RS: number of secondary on the main stem, RT: tertiary shoots on the main stem, LR: root length (cm), RC: number of lateral roots. Each seedling was then divided into roots, stems and leaves and they were dried in an oven for at least 48 h at 70°C to constant weight for biomass determination (g). PFT: Fresh stem mass, PST: dry stem mass, PFR: fresh root mass, PSR: dry root mass, RPF: root / shoot fresh mass ratio, RPS: root / shoot dry mass ratio, PFF: leaf fresh mass, PSF: leaf dry mass, FWC: leaf water content, TWC: stem water content and RWC: root water content, were determined by difference between fresh and dry mass as follows:

FWC = PFF – PSF; TWC = PFT – PST; RWC = PFR – PSR

In order to determine leaf area (SF in cm^2), the first five units were observed in April 9th (second season). Leaves were photocopied on paper, surface of 1 cm^2 of paper (P) and the image of leaf (Fi) was weighed in grams. Leaf area was calculated using the specific

Table 1. Variance analysis for stem diameter and stem length in Argan seedling grown for 14 months under five water regimes.

Source of variation	DF	Mean square	
		Stem diameter (cm)	Stem lenght (cm)
Block	3	0.017ns	205.74**
Water regime	3	5.72**	27571**
Duration of regime	10	2.732**	6783.3**
Mother-tree	7	0.457**	4027.2**
Water regime × duration	30	0.05**	172.74**
Water regime × mother-tree	21	0.095**	536.58**
Duration of regime × mother-tree	70	0.001^{ns}	7.63^{ns}
Water regime × duration of regime × mother-tree	210	0.002^{ns}	10.62^{ns}
Water regime × duration of regime × mother-tree × seedling	352	0.98^{ns}	42.18^{ns}
Error	2096	0.008	53.31

DF: Degree of freedom; ns: not significant; *: significant at 5%; **: significant at 1%.

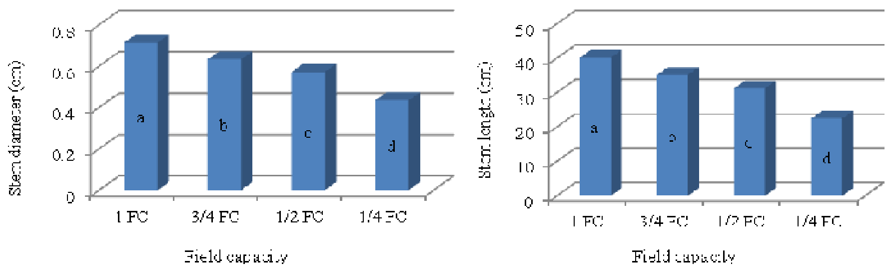

Figure 1. Stem diameter and stem length in Argan seedling grown for 14 months under five water regimes (letters indicate significant differences using LSD comparison).

surface area as proposed by Mosseddaq (1988) (S = Fi / P).

Field test

Preparation of seedlings is the same as the previous test. After germination, seedlings were grown in three types of containers measuring 17 cm in height: plastic sachets, jute sachets and seedling trays as is the case of forestry nurseries. Seedlings issued from eight genotypes grown in nursery for seven months at the Faculty of Sciences, Agadir were subjected to five water regimes [1 field capacity (1 FC = 200 ml), 3/4 FC (150 ml), 1/2 FC (100 ml) and 1/4 FC (50 ml) and 0 FC (0 ml)]. Those seedlings were then transplanted in two study plots located in the communal forests belonging to two adjacent cities: Ait Hamadi (Ouled Teima in February 2 second season) near Agadir and in Onagha near Essaouira (February 9 second season) (Water and Forests Services). Seedling height (LT) and basal diameter (DT) were recorded five times: on July 19, August 19, September 19, October 19, November 19 the second season, respectively.

Statistical analysis

In the first test, we adopt randomized complete block as experimental design. In each of the four blocks, 40 combinations (five water regime and eight varieties) are randomly distributed. Each pot is a variety and a water regime with two replications. In the second test, the experimental design is a Split-plot (Vessereau, 1988). Main water regime consists of eight mother-tree seed sources distributed in 36 plots and randomly allocated to each of the two blocks. The type of container in which Argan seedlings were grown is subsidiary water regime. Analysis of variance with four factors is adopted. All factors (block, mother-tree, water regime and observation date) were crossed. When significant differences were noted, LSD (least significant difference method) test was used to determine differences (Dagnelie, 1984). Calculations were performed using Statistix software.

RESULTS

During the pot test

Seedlings under 0 FC (field capacity) (0 ml) have dried completely after one month of stopping irrigation.

Stem characters

Water regime: Water regime is highly significant for the stem diameter and length during the test duration and at the end of the experiment (Table 1). The decrease in water amount leads to a reduction of stem diameter and stem length. Values varied respectively between 0.72 and 40.2 cm at field capacity (1 FC) to 0.43 and 22.5 cm at 1/4 FC (Figure 1).

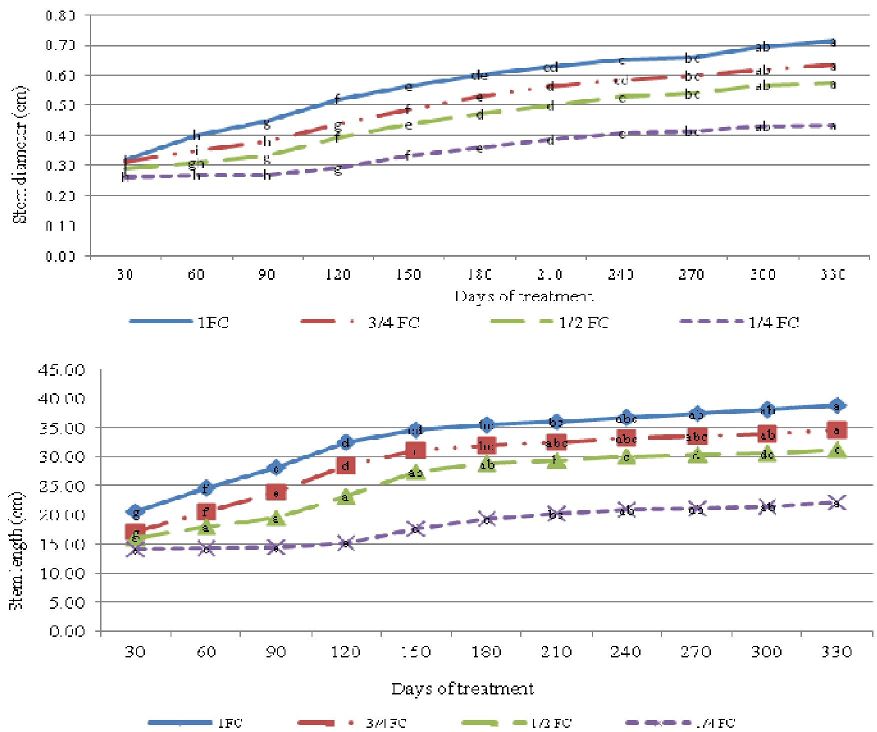

Figure 2. Stem diameter and length in Argan seedling grown for 14 months under five water regimes.

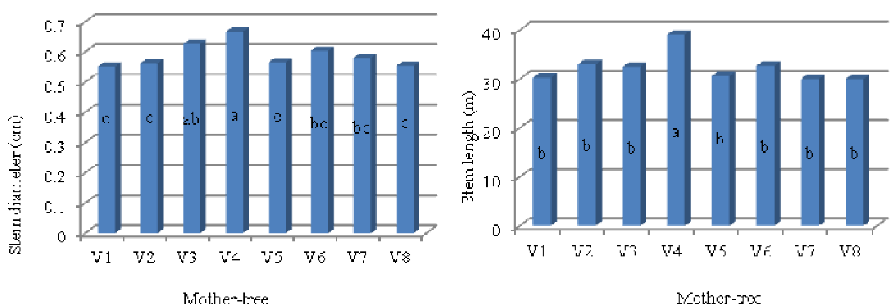

Figure 3. Stem diameter and length (cm) in Argan seedling grown for 14 months under five water regimes by mother-tree.

Duration of water regime: Duration of water regime and water regime × duration interaction influenced significantly stem diameter and length in Argan seedlings (Table 1). In seedling grown for 14 months, stem diameter remains very low at ¼ FC and ranged from 0.26 to 0.44 cm, whereas it increases in the other three water regimes and reaches 0.7 cm. Stem length increases rapidly during the first five months for water regimes 1, 3/4 and 1/2 FC and becomes slower after the fifth month (Figure 2). For 1/4 FC, stem length remains stable during the first four months, seedlings remain dwarf since length reaches only 22.5 cm at the end of the experiment.

Mother-tree: Mother-tree × water regime interaction and mother-tree were highly significant for the stem diameter and length in Argan, showing genetic variability for seedling reaction to water stress (Table 1). Diameter varied from 0.55 cm in seedlings of family V1 and V2 to 0.67 cm in family V4 (Figure 3). Stem length varied from 39.1 cm in seedlings of mother-tree V4 and 30.03 cm for V7 and V8.

At the end of experiment

Water regime

Water regime was highly significant for stem diameter

Table 2. Variance analysis for the stem, root and leaf characters in Argan seedlings grown for one year subjected to four water regime.

Source of variation	DF	Mean square: Stem characters							Mean square: Root characters				
		DT	LT	RS	RT	PFT	PST	TWC	LR	RC	PFR	PSR	RWC
Block	3	0.015^{ns}	63.32^{ns}	8.86^{ns}	3.46^{ns}	7.73^{ns}	1.74^{ns}	2.14^{ns}	1119.4^{ns}	4.80*	2.17^{ns}	0.23^{ns}	1.13^{ns}
Water regime	3	0.886**	3582.1**	12.50^{ns}	3.22^{ns}	343.1**	112.17**	63.35**	7925.6**	11.28*	966.16**	266.10**	219.17**
Mother-tree	7	0.054**	289.95**	24.02**	0.80^{ns}	44.07**	19.4**	5.14**	3824.1*	0.75*	6.56*	2.76*	1.14*
Water regime × mother-tree	21	0.016^{ns}	74.056^{ns}	11.31^{ns}	1.49^{ns}	11.07*	4.49*	1.56^{ns}	1648.1^{ns}	3.26**	10.63^{ns}	3.09^{ns}	2.55^{ns}
Water regime × mother-tree × seedling	32	0.019^{ns}	50.97^{ns}	6.98^{ns}	1.43^{ns}	5.56^{ns}	2.17^{ns}	0.82^{ns}	1312.8^{ns}	1.11^{ns}	9.76^{ns}	2.65^{ns}	2.38^{ns}
Error	173	0.014	73.27	7.71	1.67	6.32	2.41	0.97	1476.6	1.63	10.128	3.14	2.11

Source of variation	DF	Mean square: Ratios			Mean square: Leaf characters				
		RPF	RPS	RL (LR/LT)	NF	PFF	PSF	FWC	NE
Block	3	0.02^{ns}	0.04^{ns}	0.82^{ns}	10640^{ns}	1.99^{ns}	0.42^{ns}	0.78^{ns}	2247.2**
Water regime	3	2.93**	1.64**	4.64**	1654400**	146.69**	20.20**	58.09**	10514**
Mother-tree	7	2.66**	2.33*	7.61**	37321*	1.74^{ns}	0.26^{ns}	0.74^{ns}	1371.8**
Water regime × mother-tree	21	0.39*	0.31*	1.32^{ns}	64615*	1.68^{ns}	0.24^{ns}	0.77^{ns}	300.88^{ns}
Water regime × mother-tree × seedling	32	0.20^{ns}	0.17^{ns}	1.22^{ns}	34657^{ns}	1.93^{ns}	0.32^{ns}	0.77^{ns}	166.38^{ns}
Error	173	0.21	0.18	1.52	36343	1.41	0.26	0.52	274.23

DF: Degree of freedom, ns: not significant, *: significant at 5%; **: significant at 1%. Stem diameter (DT), stem length (LT), number of secondary on the main stem (RS), number of tertiary shoots on the main stem (RT), fresh stem mass (PFT), dry stem mass (PST), stem water content (TWC); root length (LR), number of lateral roots (RC), fresh root mass (PFR), dry root mass (PSR), root water content (RWC); root / shoot fresh mass ratio (RPF), root / shoot dry mass ratio (RPS), ratio length (RL = LR/LT); leaf number on the main stem (NF), leaf fresh mass (PFF), leaf dry mass (PSF), leaf water content (FWC) and spines number on the main stem (NE); Means followed by letters are significant..

and length, for spines number (EP), fresh (PFT), dry biomass weight (PST) and stem water content (TWC) at the end of experiment (Table 2). It was not significant for number of secondary, tertiary shoots on the main stem. Drought stress diminished biomass production, stems water content and reduced spines number (Figure 4).

Mother-tree effect

Water regime × mother-tree × seedling interaction was not significant for all stem traits. Mother-tree × water regime interaction was significant only for fresh and dry mass of the stem, which shows the individual response of each seedling to each water regime. Mother-tree factor was significant for all characters, except number of tertiary shoots (Table 2). Great variability was observed between all genotypes (mother-trees) for these characters. Dry mass varied from 4.5 g for mother-tree V4 to 2 g in seedlings of V1. The fresh mass in genotype V4 (7 g) was twice larger than that in seedlings from V1 (3.2 g). Stem water content varied from 2.5 g for mother-tree V4 to 1.2 g in seedlings of V1. Number of secondary shoots varied between 4.3 for mother-tree V3 to 1.5 in seedlings of V1 (Figure 5). Spines number varied from 62.6 in mother-tree V4 and 42.9 in seedling from mother-tree V1.

Root characters

Water regime

Water regime is highly significant for root length (LR), number of lateral roots (NRL), fresh (PFR), dry mass (PSR), root water content (RWC), fresh

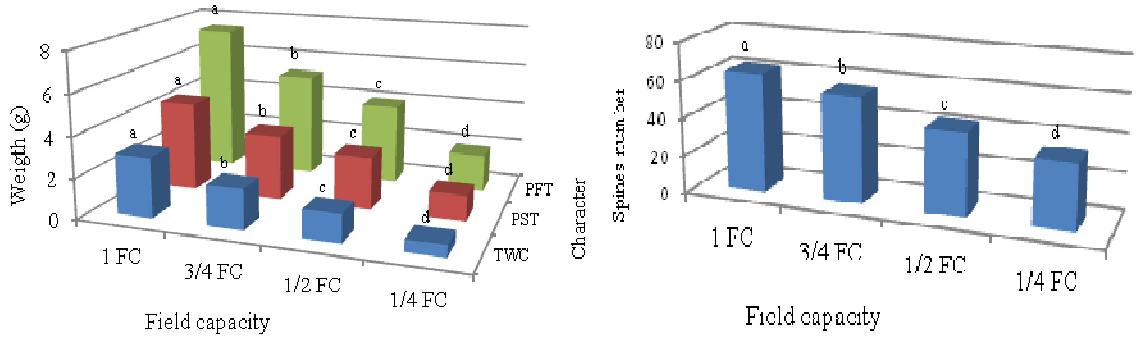

Figure 4. Fresh, dry weight and stem water content and spines number in Argan seedlings grown for 14 months by water regime.

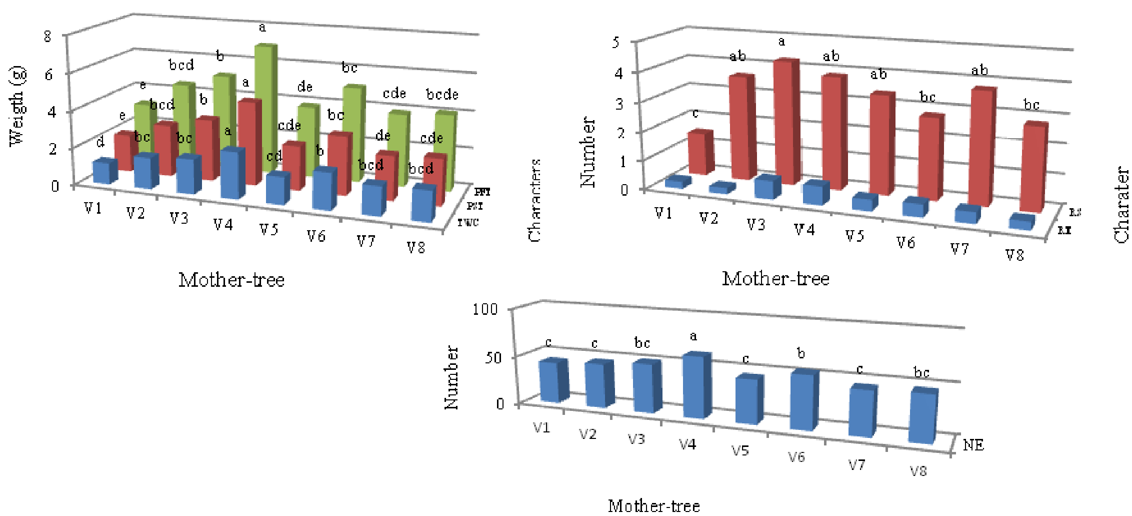

Figure 5. Fresh, dry mass, water content and number of secondary, tertiary shoots and spines on the main stem by mother-tree.

(RPF) and dry (RPS) mass ratio and length ratio (RL) (Table 2). Root length varied from 49.9 cm at ¼ FC to 73.4 cm at 1 FC, it is about twice the stem length (Figure 6). The decrease in the amount of irrigation water leads to a reduction in lateral roots number, root water content and root fresh mass. The fresh mass is at field capacity six times greater decreases also than that obtained in ¼ FC. Root dry biomass at field capacity is five times larger than that recorded at ¼ FC. The fresh and dry weight ratios varied from 1.7 and 1.6 at field capacity to 1.1 and 1.2 at 1/4 FC. Root / stem ratio of length (RL) varied from 2.4 at ¼ FC to 1.9 at 1 FC.

Mother-tree

Mother-tree × water regime was not significant for all root characters except number of lateral roots. This shows genetic variability in Argan seedling for water stress. Mother-tree × water regime × seedling interaction is not significant for all root traits. The production of root fresh, dry biomass and water content depends only on water regime. Mother-tree factor was significant for root length (LR), root / stem ratio of length (RL), fresh (RPF) and dry (RPS) mass ratio (Table 2). Thus, seedlings from genotype V1 have longer roots (82.3 cm) than seedlings from V4 which shows the shortest roots (53.13 cm) (Figure 7). Water stress induces the development of root to allow the seedling in search of water in depth. Thus, root / stem ratio varied from 2.9 in seedlings from mother-tree V1 to 1.4 in descendants of mother V4. Fresh and dry mass ratio varied from 2 and 1.9 in seedlings from V1 to 0.92 and 0.89 for descendants of V4.

Leaf characters

Water regime

Water regime is highly significant for leaf number on the

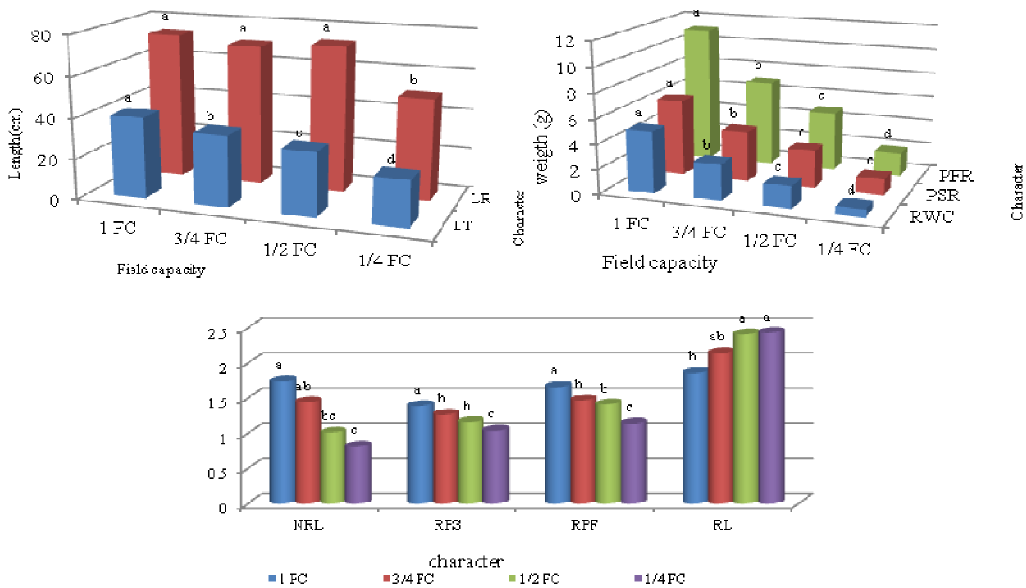

Figure 6. Stem and root length, fresh, dry mass and root water content and their ratios in Argan seedlings by water regime.

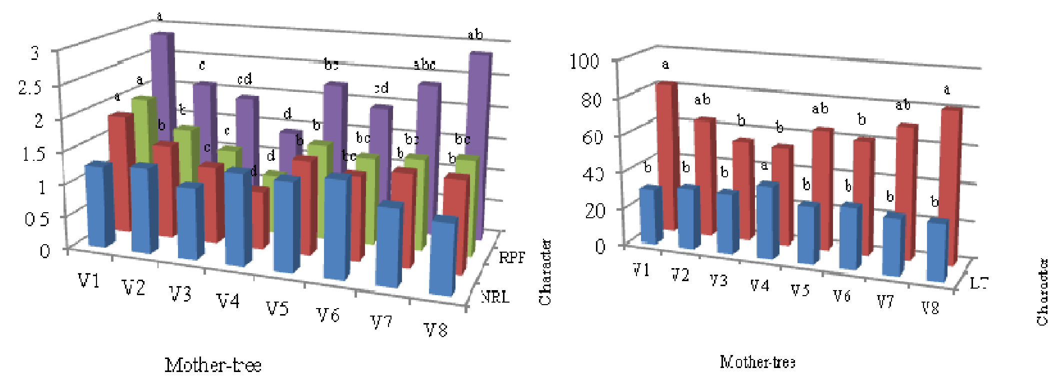

Figure 7. Root and stem length, root and stem ratio of length, fresh, dry mass and number of lateral roots in Argan seedlings by mother-trees.

main stem (NF), fresh, dry mass and leaf water content (Table 2). At field capacity, leaf number (584.6) is three times higher than that obtained at 1/4 FC (201.8) (Figure 8). Water stress induces a significant reduction of leaf biomass, since fresh weight at field capacity (4 g) was eight times greater than that obtained at 1/4 FC (0.5 g). Leaf dry mass does not exceed 0.3 g at 1/4 FC while it reaches 1.6 g at field capacity. Water content at field capacity (2.4 g) is twelve times greater than that obtained at 1/4 FC (0.2 g).

Mother-tree

Water regime × mother-tree × seedling interaction was not significant for leaf characters. Mother-tree and water regime × mother-tree were significant for leaf number but not significant for the three other characters (Table 2). Leaf number varied between 437.1 in seedlings of V4 and 319.3 in seedlings of V5. Fresh and dry biomass varied from 1.7 g, 0.78 g in seedlings of V5 and 2.36 g, 1.01 g in seedlings of V3. Leaf water content varied from 0.91 in seedlings of V5 and 1.34 in seedlings of V3 (Table 3).

Field test

Before transplanting

Seedling, mother-tree × seedling, water regime × seedling and water regime × mother-tree × seedling were

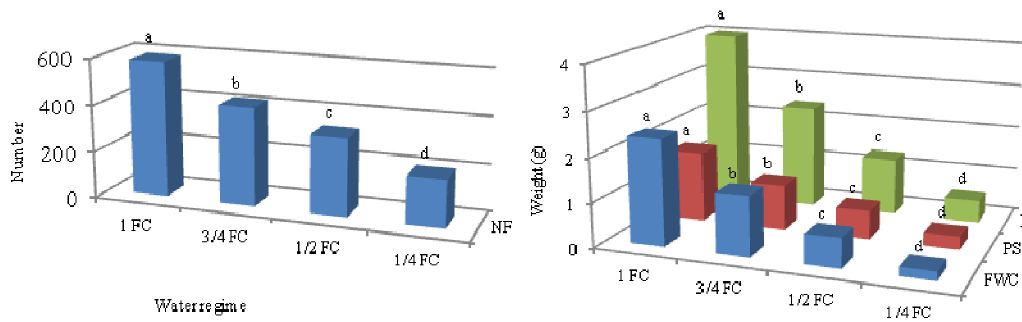

Figure 8. Leaf number on the main stem, fresh, dry mass and water content by water regime.

Table 3. Fresh (PFF), dry mass (PSF), leaf water content (FWC) and leaf number by mother-tree.

Mother-tree	PSF	FWC	PFF	NF
V1	0.82	0.99	1.81	380.4^{bc}
V2	0.95	1.16	2.11	413.1^{ab}
V3	1.01	1.34	2.36	379^{bc}
V4	1.01	1.29	2.3	437.1^{a}
V5	0.78	0.91	1.7	319.3^{d}
V6	0.88	1.26	2.14	388.1^{b}
V7	0.81	1.06	1.87	369.4^{c}
V8	0.85	1.21	2.06	390.4^{b}

Means followed by letters are significant.

Table 4. Variance analysis for seedling height (LT), basal diameter (DT) under five water regimes grown in three containers for seven months.

Source of variation	DF	Average	
		DT	LT
Block	3	0.005^{ns}	2.09^{ns}
Mother-tree	7	0.029*	330.95**
Water regime	4	0.56*	67.77*
Seedling	1	0.007^{ns}	25.34^{ns}
Water regime × mother-tree	28	0.005^{ns}	19.04*
Mother-tree × seedling	7	0.003^{ns}	4.02^{ns}
Water regime × seedling	4	0.001^{ns}	2.43^{ns}
Water regime × mother-tree × seedling	28	0.003^{ns}	7.11^{ns}
Error	236	0.004	10.79

ns: not significant; *: significant at 5%; **: significant at 1%.

not significant for stem diameter (DT) and stem length (LT). Water regime × mother-tree is significant for LT but not significant for DT. Mother-tree and water regime were significant for the two characters of seedlings grown for seven months under five water regimes in three containers (Table 4). Water stress causes a decrease in DT from 0.45 cm at field capacity to 0.21 at 0 FC. Stem length varied from 16.15 cm at 1 FC to 13.44 cm at ¼ FC (Table 5). Stem length and diameter varied in large proportions between mother-trees. Values ranged respectively between 0.29, 10.92 cm in seedlings of V8 and 0.37, 20.22 cm in seedlings of V4 (Table 6). Great variability in leaf area was observed between mother-trees and between water regimes for the same mother-tree. Values varied between 0.43 cm^2 in seedlings of V7 at 1 FC and 1.37 cm^2 at 0 FC in seedlings of V3 (Table 6).

After transplanting

Water regime was significant for stem diameter (DT), but

Table 5. Seedling height (LT) and stem diameter (DT) in Argan seedlings grown for seven months under five water regimes.

Water regime (FC)	Average	
	DT (cm)	LT (cm)
1	0.45[a]	16.15[a]
¾	0.38[b]	14.44[b]
½	0.33[c]	14.24[b]
¼	0.26[d]	13.44[b]
0	0.21[e]	13.90[b]

Means followed by letters are significant.

Table 6. Seedling height (LT) and stem diameter (DT) in Argan seedlings grown for seven months under five water regimes by mother-tree.

Mother-tree	Average	
	DT	LT
V1	0.31[cd]	12.49[cd]
V2	0.3[cd]	14.17[c]
V3	0.36[ab]	16.71[c]
V4	0.37[a]	20.22[a]
V5	0.31[cd]	14.22[c]
V6	0.33[bc]	13.97[c]
V7	0.32[cd]	12.76[cd]
V8	0.29[d]	10.92[d]

Means followed by letters are significant.

Table 7. Variance analysis for stem diameter (DT), stem length (LT) in Argan seedlings transplanted in two study plots (Ouled Teima and Essaouira).

Source of variation	DF	Mean square	
		LT (cm)	DT (cm)
Block	3	35.96[ns]	1.1[ns]
Mother-tree	7	2071.5**	1.31[ns]
Water regime	4	1125.7**	4.96**
Observation date	4	3769**	1.23[ns]
Mother-tree × water regime	28	178**	1.08[ns]
Mother-tree × observation date	28	3.8[ns]	1.03[ns]
Water regime × observation date	16	455.73**	1.06[ns]
Mother-tree × water regime × observation date	112	7.73[ns]	1.02[ns]
Error	1387	30.5	1.04

Ns: Not significant, **: significant at 1%.

mother-tree, observation date, mother-tree × water regime, mother-tree × observation date and mother-tree × observation date × water regime interactions were not significant for this character. Water regime, mother-tree, observation date, mother-tree × water regime, water regime × observation date interactions were significant for stem length (LT). Mother-tree × observation date and mother-tree × water regime × observation date interactions were not significant for this character (Table 7). The reducing of water availability causes a reduction of stem length since values are from 28.03 cm at field capacity to 14.08 cm at 0 FC. Stem diameter varied from 0.52 cm at ¾ FC and 0.21 cm at 0 FC (Table 8). Stem diameter and length varied in large proportions between families. Values ranged respectively between 0.3 cm, 17.44 cm in seedlings of V8 and 0.38 cm, 27.27 cm in seedlings of V4 (Table 9). Values reached in seedlings in field test remained lower than pot test even when they

Table 8. Stem length (LT), stem diameter (DT) in Argan seedling of eight mother-trees grown in field and subjected to five water regime.

Water regime (FC)	Average	
	LT (cm)	DT (cm)
1	28.03[a]	0.45[ab]
¾	24.17[b]	0.52[a]
2/4	20.8[c]	0.35[bc]
¼	15.09[d]	0.28[c]
0	14.08[e]	0.21[c]

Values followed by letters are significantly different.

Table 9. Stem length (cm) and stem diameter (cm) by mother-tree having undergone five water regimes.

Mother-tree	Average	
	LT (cm)	DT (cm)
V1	17.55[d]	0.32[c]
V2	20.29[c]	0.31[c]
V3	22.44[b]	0.37[a]
V4	27.27[a]	0.38[a]
V5	20.04[c]	0.33[bc]
V6	19.9[c]	0.35[b]
V7	18.43[d]	0.33[bc]
V8	17.44[d]	0.3[c]

Values followed by letters are significantly different.

were grown for the same period about 14 months (Table 10). Seedling, water regime × mother-tree, mother-tree × seedling, water regime × seedling and water regime × mother-tree × seedling were not significant for leaf area (SF). Mother-tree and water regime were significant for leaf area in Argan seedlings grown for seven months and subjected to five water regime (Table 11).

Leaf area decrease with water deficit, it varied between 0.71 cm^2 at 1 FC and 0.66 cm^2 except at 0 FC (0.76 cm^2) (Table 12). Some genotypes as V3 and V4 have greater leaf areas than seedlings from V7, V8 and V1.

DISCUSSION

The mechanisms adopted by Argan seedlings to withstand drought are still largely unknown. Here, we report data of experiments aimed at investigating the effect of water stress on Argan seedlings growth issued from eigth mother-tree originating from three populations in south west Morocco. In our study, the growth reduction in both seedling height and basal diameter of the stem was observed under moderate (3/4 FC) and severe stress (1/4 or 0 FC). The same result is obtained by Kaabouss (1992) since the stem length of Argan seedlings subjected to water stress did not exceed 22 cm after 160 days of transplantation. In seedlings of *Adenanthera pavonina*, Paliwal and Kannan (1999) showed that water deficit causes a reduction of the stem height. This result suggests that even at reduced soil water availability, the Argan seedlings are able to grow. Our results agree with the findings of Achten et al. (2010) in *J. curcas* since under medium stress (40% plant available water) the plants maintained a similar stem shape, although they grew at lower rate (stem length: 0.28 cm / day; dry biomass production: 0.64 g / day). Seedlings under extreme drought stress (no irrigation) stopped growing, started shedding leaves and showed shrinking stem diameter from the 12th day after the start of the drought treatment. This result showed that water withhold would arrest growth but maintaining plants at low soil water availability (40%) would allow them to continue growing, although at a slower rate than fully irrigated plants (Achten et al., 2010; Sapeta et al., 2013).

Decrease in irrigation level leads to a low level of shoot (secondary and tertiary shoots) and spines production which agree results obtained in adult trees since branching is very low in dry season than in wet season. In addition, the production of spines is not a response to drought (Zahidi, 1997). Drought stress leads also to the decrease of stem fresh mass, dry mass and stem water content in argan seedlings at the end of experiment.

Table 10. Stem length (cm) and stem diameter (cm) in transplanted Argan seedlings grown under five water regimes.

Observation date	Average	
	LT (cm)	DT (cm)
July 19	16.3^{e}	0.27
August 19	18.27^{d}	0.43
September 19	20.01^{c}	0.32
October 19	22.67^{b}	0.37
November 19	24.92^{a}	0.4

Values followed by letters are significantly different.

Table 11. Variance analysis for leaf area (SF) in Argan seedlings transplanted in two study plots (Ouled Teima and Essaouira).

Source of variation	DF	SF
Block	3	0.34^{ns}
Mother-tree	7	6.43**
Water regime	4	0.65*
Seedling	1	0.04^{ns}
Water regime × mother-tree	28	0.53^{ns}
Mother-tree × seedling	7	0.37^{ns}
Water regime × seedling	4	0.11^{ns}
Water regime × mother-tree × seedling	28	0.33**
Water regime × mother-tree × seedling × leaf (error)	1506	0.19

Ns: Not significant, *: significant at 5%, **: significant at 1%.

Table 12. Variation of leaf area by mother-tree and water regime.

Mother-tree	1 FC	3/4 FC	2/4 FC	1/4 FC	0 FC	Average
V1	0.63^{ab}	0.67^{a}	0.64^{ab}	0.56^{bc}	0.48^{c}	0.59^{cd}
V2	0.71^{a}	0.54^{b}	0.51^{b}	0.64^{a}	0.54^{b}	0.58^{cd}
V3	1.07^{ab}	0.82^{b}	0.77^{b}	0.99^{ab}	**1.35**^{a}	1.03^{a}
V4	0.93^{a}	0.88^{b}	0.85^{c}	0.87^{b}	0.96^{a}	0.89^{ab}
V5	0.79^{a}	0.64^{c}	0.77^{ab}	0.66^{bc}	0.8^{a}	0.73^{b}
V6	0.61^{cd}	0.64^{c}	0.91^{a}	0.51^{d}	0.84^{b}	0.7^{bc}
V7	**0.43**^{d}	0.48^{cd}	0.61^{a}	0.51^{bc}	0.57^{ab}	0.52^{d}
V8	0.55^{a}	0.46^{b}	0.45^{b}	0.57^{a}	0.51^{ab}	0.5^{d}
Average	0.71^{ab}	0.64c	0.68b^{c}	0.66b^{c}	0.76^{a}	0.69

Values followed by letters are significantly different.

Drought stress obviously diminished the biomass and their components, and reduced shoot percentages in *S. davidii* seedlings (Wu et al., 2008). Additionally, photosynthesis and growth (biomass production) are the primary processes to be affected by drought (Chaves and Oliveira, 2004; Sapeta et al., 2013). Drought not only changed plant growth and structure [shoot height (Ht), total biomass (Tb)] in *P. davidiana*, but also affected plant physiological properties and constitutes a very important limiting factor at the initial phase of seedling growth and establishment (Zhang et al., 2004). Drought changed also root growth, root to shoot length which is twice higher under sever stress (at 1/4 FC). Thus, water stress induces the development of root to allow the seedling in search of water in depth.

The decrease in the amount of irrigation water leads to a reduction in lateral roots number, fresh, dry mass, root water content, root to shoot fresh and dry mass ratio. Our results agree with the finding in other species, Wu et al. (2008) showed that drought stress diminished the root biomass and their components, but increased below-ground percentages. Drought stress significantly decreased plant total dry mass, but the proportion of changes differed among root and stem. The change in root to shoot dry mass ratio has been considered as one of the mechanisms involved in the adaptation of plants to drought stress (Turner, 1997). So, in order to diminish consumption and increase absorption of water, plants in dry conditions often decrease their growth rate and biomass production, and contribute more biomass to roots (Martin and Stephens, 2006; Wu et al., 2008).

Additionally, the importance of root systems in acquiring water has long been recognized as crucial to cope with drought conditions in acquiring water. A prolific root system can confer the advantage to support accelerated plant growth during the early growth stage after transplantation and extract water from shallow soil layers that is otherwise easily lost by evaporation especially in arid environments (Johansen et al., 1994). Water stress induces a significant reduction of leaf number, leaf biomass (fresh, dry mass) and leaf water content in Argan seedlings. In general, under drought stress (3/4, ½ and ¼ FC), all genotypes had lower values of leaf area than the well-watered regime (1 FC) except at 0 FC. However, drought resulted in a reduction of total leaf area in seedlings of several species. Reduced leaf number and area was a drought avoidance strategy for the seedlings by reducing transpiration. These results confirm the conclusion that drought constrains leaf growth to a much greater extent, indicating that this abiotic factor led to a more conservative balance between losing and obtaining water by different organs of plant (James and William, 1998; Liu and Stützel, 2004; Zhang et al., 2004; Villagra and Cavagnaro, 2006; Wu et al., 2008). Some genotypes as V4 and V3 unlike V1 and V8 are able to grow in length and diameter, able to produce more shoots, spines and biomass (fresh mass, dry mass) and with much water content in the stem even under water stress. These results suggest the existence of mechanisms of drought tolerance in these genotypes.

As proposed by Liu and Stützel (2004), conservative shoot growth during drought could be advantageous, especially if root growth is promoted. Genotypes that sustain shoot growth during drought may have greater marketability, which is particularly important for leafy vegetable crops. In Argan, under drought, genotype V4 and V3 have taproots exceeding 50 cm, which are accompanied by a large number of lateral roots, more leaves, a large biomass (fresh, dry mass and water content) and great leaf areas even under severe stress (1/4 and 0 FC). Those genotypes are promising for reforestation programs in an environment where drought is a limiting factor for the natural regeneration. Therefore, the most difficult in vegetation growth in this area appears in summer for newly planted seedlings, their survival or death will depend on adaptation to the habitat through this crucial period as reported for four shrub species in arid valley of Minjiang River of China (Li et al., 2007). Genotypes (mother-tree of seedlings) V1 and V8 have produced longer roots, higher root to shoot ratio and root biomass production (fresh, dry weight ratios and water content), lower leaves number, a relatively lower biomass (fresh, dry mass and water content) and smaller leaf areas than seedlings from genotypes V4 and V3. In many species,root system in acquiring water has long been recognized as crucial to cope with drought conditions (Kashiwagi et al., 2006).

The increased root (root length, root biomass production) and leaf characters (biomass and area) under drought stress observed in genotypes V1 and V8 is in line with the theory of the functional balance, which predicts that plants will react to a limited water availability with a relative increase in the flow of assimilates to the root leading to an increased root biomass. So, in order to diminish consumption and increase absorption of water, plants in dry conditions often decrease their growth rate and biomass production, and contribute more biomass to roots (Villagra and Cavagnaro, 2006). Partitioning more biomass to below-ground and maintaining higher root to shoot ratio may be beneficial to enhanced capacity of water uptake, by maintaining the shoot in a well-hydrated condition (Blum, 1996; Liu and Stützel, 2004; Zhang et al., 2004; Kashiwagi et al., 2006; Wu et al., 2008). At this stage of seedlings development, this case will be an adaptative response of the plant morphology which may be a primary mechanism by which this species can cope with the environmental characteristics of south west Morocco as reported for other species (Patterson et al., 1997; Achten et al., 2010; Sapeta et al., 2013).

Conclusion

The Argan seedlings grown in pot and field showed that water stress was a very important limiting factor at the initial phase of growth. Drought stress significantly decreased seedling height, root growth, leaf area and biomass production, but the proportion of changes differed among root, stem and leaf. Great genetic variability between seedlings to water stress. Even under severe stress, some genotypes increased root to shoot fresh and dry mass ratio and maintained higher investment in the taproot and lateral roots. This adaptive response in morphology may be a primary mechanism by which the Argan seedlings can cope with the environmental characteristics. For newly planted seedlings, the most difficult period for plants growth in this area appears from May to October since rainfall not exceeds 40 mm. So, in order to enhance the survival rate and conserving rate of seedlings planting, appropriate human intervention is required to reduce the damage to Argan seedlings resulting from drought. These results will be useful towards transplantation, and may serve as a guide to initiate effective measures in tree planting to enhance survival rate and conserving rate for Argan forest restoration.

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