Volume 75(5). Published September, 01, 2020
www.jnsciences.org
E-ISSN 2286-5314

Morphological diversity of the afares oak (Quercus afares) compared to the cork oak (Quercus suber) and the zeen oak (Quercus canariensis) in Kroumirie (Aïn Zana)

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Abstract - By its geographical position, Tunisia presents a great diversity of biotopes occupied by an important floristic richness presenting the various plant species. It includes among others the genus Quercus which is represented by five species, two of which are deciduous (zeen oak and afares oak) and three sclerophyll (cork oak, kermes oak and holm oak).
A morphological study of afares oak compared to zeen oak and cork oak was carried out under semi-controlled conditions (in the nursery) where the results show a higher germination rate in afares oak (96%), a morphological variability of acorns and leaves increase in height in cork oak (12.04 cm) compared to zeen oak (8.76 cm) and afares oak (5.1 cm). Under natural conditions (on the ground) dendrometric measurements carried out on adult trees show that the height is greater in afares oak and zeen oak (about 15 m) and to a lesser degree in cork oak (about 10 m) while the leaves showed a great morphological variability except for the cork oak and the afares oak where the latter constitutes a forest species of intermediate form between the two oaks mainly due to the mix of stands in the station.
The afares oak species differs from other acorn size oak species, especially with cork oak as well as the increase in height and diameter (young plants and adult trees) as well as the size of leaves. All these parameters of afares oak are close to that of zeen oak and cork oak at the same time while retaining an intermediate form between them and admitting the idea of hybridism.

Keywords: Quercus afares, Quercus canariensis, Quercus suber, germination, morphological parameters.

1. Introduction

In the future, the oak forest is therefore seriously threatened by destructive actions causing an increasing reduction in wooded areas, thereby reducing their protective and socio-economic roles, especially with regard to the afares oak (*Quercus afares*) located in the Mediterranean basin by comparing it with cork oak (*Quercus suber*) and zeen oak (*Quercus canariensis*).
Mediterranean oak groves are characterized by the presence of deciduous oaks (zeen oak) and evergreen oaks (cork oak and kermes oak) but a study of the distribution of oak groves in north-western Tunisia proves that the afares oak represents a systematic morphology comparable to that of cork oak and zeen oak. They are a broad-leaved forest species comparable to a tree-like habit that is most often found at different sites such as El Feidja, Djebel El Ghorra and Aïn Zena. Its geographical area extends from West Kroumirie in Tunisia to Cherchell in Algeria.
we take this study research in order to contribute a better knowledge of the origin of this oak as being a hybrid species resulting from cork oak and of zeen oak to better understand its natural way of life and its degree of resistance to environmental constraints in order to conserve its natural and artificial regeneration in Tunisia given their plasticity and resistance to extreme conditions, as well as their socio-economic and ecological value.
In fact, it is very important to study the morphological diversity of afares oak by comparing it with zeen oak and cork oak in Kroumirie and to expose all the quantitative and qualitative results obtained under natural cultivation conditions (on field) and in semi-controlled conditions (in the nursery); A comparative study of young plants in semi-controlled conditions (in the nursery) and adult trees in natural conditions (in the field).

2. Materials and methods

The study focused on acorns, young plants and adult trees of afares oak (*Quercus afares*), cork oak (*Quercus suber*) and zeen oak (*Quercus canariensis*) in the site of Ain Zana (Ain draham).

The experiment was carried out under semi-controlled conditions in experimental apparatus in I.N.R.G.R.E.F-Ariana (upper semi-arid bioclimate). The acorns of afares oak, zeen oak and cork oak were harvested in November and December in the Ain Zana station. They were sorted and kept in a cold room at a temperature equal to 4 ° C for a week, and then they were sown, without any pre-treatment. Sowing of afares oak, zeen oak and cork oak acorns which was carried out in November and December, without any prior treatment, in perforated polyethylene bags 16 cm deep and 10 cm in diameter filled with a mixture of sand and oak humus with a weight of about 1.7 Kg (one acorn per sachet) in order to install young plants of afares oak, zeen oak and cork oak.

The soil moisture at field capacity of the substrate is around 26%: **(Tab 1)**

Table 1: Soil texture study (in semi-controlled conditions)

Material	Clay %	Silt %	Fine sand %	Sand coarse %	Lime stone Total %	OM %	pH	P_2O_5 %	K_2O %
	12	12	30	43	3	3.2	7.5	4	0.17

2.1 Study of germination

Germination is the resumption of the active life of a plant after a period of rest, of variable duration. The environmental conditions (humidity, temperature) coupled with the characteristics of the seed (thickness of the seed coat, physiological mechanism, conservation) that determined the germination conditions.

We followed the kinetics of germination by noting the number of acorns to determine the maximum germination rates during four months (starting from sowing date).

2.2 Measured parameters

2.2.1 Characterization of acorns

Length and diameter measurements were made on the tassels using a caliper, as well as weight measurements using a precision balance.

2.2.2 Germination rate

We followed the germination rates of the acorns of *Quercus afares*, *Quercus suber* and *Quercus canariensis*; it is the rate of emergence of acorns which corresponds to the percentage of these acorns to give seedlings.

2.2.3 Study of the growth of young plants

The morphological quality of the plants is of great importance because it offers maximum guarantees of recovery and good start of the young plantation.

2.2.4 Rod height and collar diameter

These two parameters are considered among the morphological factors that can best predict the performance of plants after planting and growth in height.

The diameter at the collar is the best predictor of plant survival (Mexal, 1990). It is generally correlated with different morphological parameters (height, dry matter weight of the roots and the aerial part), because it is an integrating parameter of the morphological response to environmental factors. The diameter at the collar can explain more than 97% of the variation observed in the total weight (Van Den Driessche, 1982).

2.2.5 Study of the morphological variability of the leaves

We collected leaves (those from the base, middle and end) from the three species of afares oak, zeen oak and cork oak (four months old); the parameters which was studied in this work are the size (length, width and length of the petioles) and we analyzed the different shapes (oval, rounded, etc.) and the leaf area (measured using a planimeter; Δt Area meter, Δ t Devices).

2.2.6 Natural conditions)

Our study focused on mature trees of afares oaks, zeen oaks and cork oaks and took place in the region of Kroumirie where we chose a study area taking into account climatically conditions, flora and altitude ;This is Ain Zena station (plot 20), located in the Ain Draham area.

2.2.7 Characteristics of Ain Zana station

The forest of Ain Zena is approximately a rectangle and occupies a fairly massive shape with an elongation along the west axis with a maximum length of 12.5km. .

The forest takes its name from the source of Ain Zana, which located at 30 km of the south of Ain Draham and South East exposure, with an area of 47 ha and is located near 1015 m above sea level. It is located in Southeast of Ain Draham in the Jendouba Governorate. (DGF,1986).

From a climatic point of view, it is part of the superior humid bioclimatic stage. The soils are mulls on sandstone and clay (Hasnaoui, 1986). Precipitation and temperatures are comparable to those recorded at Aïn Draham.

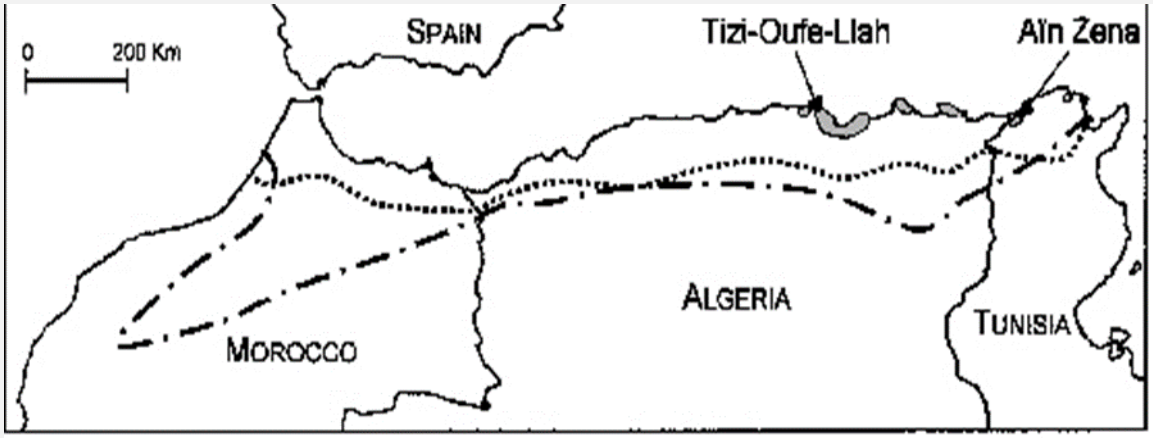

Figure 1: Geographical distribution map of the study site: Aïn Zana

2.2.8 Soil analysis

We then tried to know a certain number of information (description, structures and main types) on the soil on which the tree was studied in order to characterize the stations which based on an edaphic point of view. Therefore, a completed and detailed study of a soil profile was carried out in a homogeneous area representative of the average situation of the station. This pedological characterization study has two aspects (Duchaufour, 1965): a morphological description of the profile in the field and a summary analysis of the soil in the laboratory in order to determine the pH, texture, carbon, total nitrogen, total organics' matter, humic matter (fulvic acids + humic acids), total iron, free iron, as well as the saturation rate and the conductivity of the soil.

Soil samples taken from soil profiles dug in the Ain Zana station were subjected to physico-chemical analyzes. These were taken systematically between 0 and 10 cm for the surface horizon and between 10-30 cm and 30-50 cm for the subsurface horizon. The choice of these horizons is justified by the fact that their roles are essential in the installation of the seedling and its evolution during the first period of its life.

2.3 Study of measured parameters

2.3.1 Measurement of physical soil parameters

- The grain size (texture) is made by the Robinson pipette method, which consists in determining the quantity of the silty fractions, sandy fractions and clay fractions. This separation technique obeys the Stocks law, according to which the fall of suspended solid particles is a function of their diameter.
- Total limestone: It is measured using Bernard's calcimeter. By determining the calcium carbonate (Ca CO_3) by hydrochloric acid (HCl); These are considered that the total limestone presents almost all of the carbonate.
- Determination of carbon and organic matter: The determination of carbon is carried out by Walkey-Black methods; based on the oxidation of organic carbon by potassium dichromate in an acid medium. The value of organic matter is obtained by multiplying the carbon value (C%) by the coefficient calculated for cultivated soils, which is equal to 1.72: **M.O (%) = C (%) 1.72**

2.3.2 Measurements of chemical parameters

- Electrical conductivity (CE): Express the content of soluble salts in a solution; the measurements are carried out using a conductimeter, in a soil / water solution = 1/5, at a reference temperature equal to 20° C.

- pH (hydrogen potential): The pH is measured on a suspension of fine earth. The liquid / earth weight ratio must be constant (2.5 for example). The pH must be taken first in distilled water and then in KCl (4N) solution; using the electronic method at the glass electrode.

Profile analysis

Table 2: Physico-chemical characteristics of the soil at Aîn Zana station

Elements	Profile	P1			P2			P3
	Depth	0-10cm	10-30cm	30-50cm	0-10cm	10-30cm	30-50cm	0-50cm
Clay (%)		26	30	14	47	29	31	34
Fine silt (%)		30	17	26	16	31	29	27
Coarse silt (%)		13	12	9	14	14	13	13
Fine Sands (%)		20	25	21	13	17	17	17
Coarse Sands (%)		10	12	28	9	9	8	8
PH (1/2.5)		6.2	6.7	6.6	6.2	6.9	7	6.8
Saturation (ml/100g)		59	52	62	73	51	51	50
Conductivity (mmho/cm)		0.8	0.5	0.6	0.8	0.5	0.6	0.5
Total lime stone (%)		3	4	3	5	2	2	5
Organic material (%)		3.7	1.9	0.8	6.1	2.7	2	2.4
Carbone (%)		2.1	1.1	0.4	3.5	1.6	1.2	1.4

In general, all the profiles (P) observed are characterized by a layer of litter on the soil surface which is in the form of a thin horizon reaching, and sometimes, exceeding 5 cm. This horizon is generally followed by a blackish horizon which is rich of organic matter resulting from rapid mineralization of the brown forest; it varies depending on the plant cover. Then, a succession of horizons appears reflecting the pedogenetic nature of the profile.

The particle size analysis of the profiles (texture triangle) shows that:

P1: The horizon 0-10 cm is of clayey silt texture; the horizon 10-30 cm and 30-50 cm are of sandy clay texture and have an acidic PH.

P2: for the depth 0-10 cm, the texture is clay loam; the same for the 10-30 cm and 30-50 cm soil layers with an acidic pH which becomes more and more neutral within 0-30 cm soil layer.

P3: this profile presents the same type of soil for the entire 0-50 cm depth which it is clayey silt texture and has an acidic pH.

The organic matter decreases in all profiles with depth, this decrease can be explained by the lack of incorporation of organic matter in depth.

2.3.3 Measurements of tree dendrometric parameters

The study of the growth of afares oak, zeen oak and cork oak is very important, because it allows us to know the age corresponding to the determination of dismantling and the number of harvests to be expected. The morphological approach is based on determining the height and diameter of ten trees selected from the Ain Zana site;

The height of the trunks and the circumference of the trees (1.30 m) are measured using a tape measure. The climatic conditions of the measurement days are summarized in Table 9. The light intensities ranged from 105.6 to 161.6 µmol m-2 s-1 and temperatures fluctuated from 13.17°C to 21.86 °C and the relative air humidity is around 65%.

2.3.4 Experimental apparatus

Under semi-controlled conditions, the experimental device consists of three blocks which each one has a capacity of 100 sachets, or 100 plants per species; afar oak, zeen oak and cork oak (four months old)

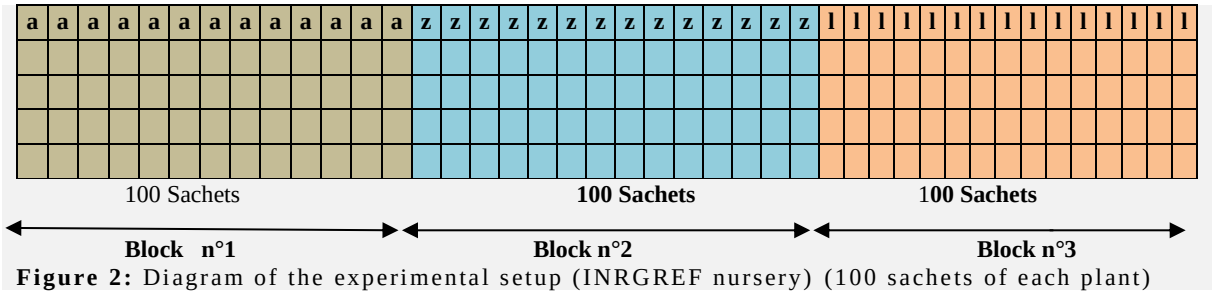

Figure 2: Diagram of the experimental setup (INRGREF nursery) (100 sachets of each plant)

Block n ° 1: afares oak Block n ° 2: zeen oak Block n ° 3: cork oak

Under natural conditions, ten adult trees of each species (afares oak, zeen oak and cork oak) from the Ain Zana station were used for the measurement of dendrometric parameters (heights and diameters). In this site, all the selected trees are identified and numbered in a logical order using paint.

2-4 Statistical analysis

Statistical analysis was carried out on the parameters (morphological factors) which are measured and obtained either under semi-controlled conditions or under natural conditions. We used the STATITCF software (version.V), (Beaux, 1991).

All the measures were analyzed by one or two factor variance depending on the case using Fisher's F test to test the hypothesis of equality of means at the 5% risk threshold. It is supplemented by multiple comparisons of means by the Newman and Keuls test when the hypothesis of equality of means is rejected, according to (Steel Robert, 1980). The graphic outputs were produced with Excel XP software.

3. Results and discussion

Under semi-controlled conditions (Nursery), the study of the morphological variability of the acorns shows that the average values of the length, diameter and weight of acorns are: 4.09 cm and 1.72 cm and 7.22 g; 3.80 cm and 1.62 cm and 6.94 g; 2.93 cm and 1.56 cm and 6.65 g respectively for the afares oak, the zeen oak and the cork oak. (Fig. 3 and 4). Statistical analysis shows a significant difference between the species by the Fisher F test at the 5% threshold.

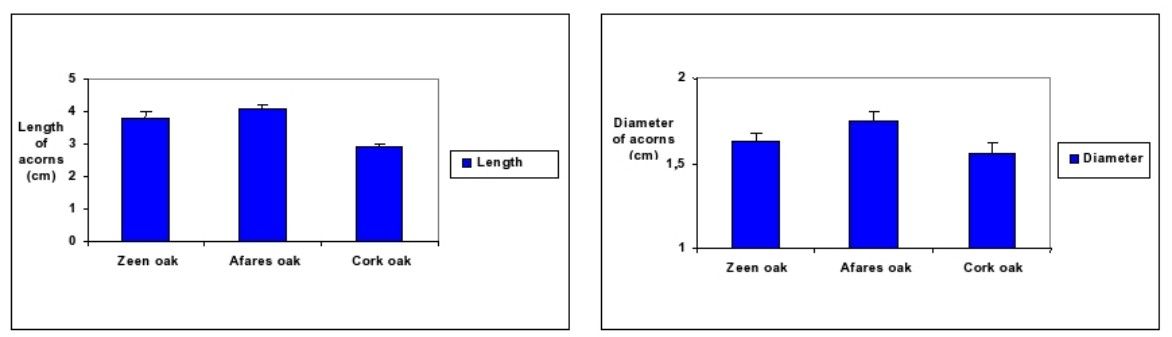

Figure 3: Length (cm) and diameter (cm) of acorns of afares oak, zeen oak and cork oak in Aïn Zana

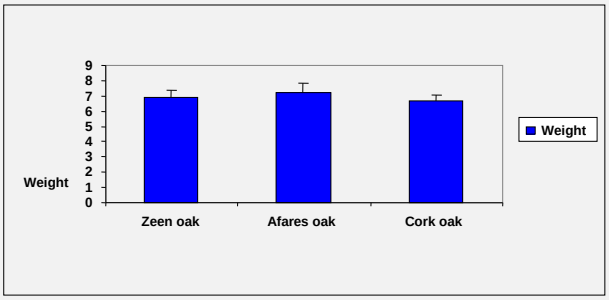

Figure 4: Weight of acorns (g) of afares oak, zeen oak and cork oak in Aïn Zana

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Cups: A description of the cup (smooth, pointed or hooked) made to each acorn and to each species shows that the afares oak and the cork oak have a hooked cup while that of zeen oak is pointed. After 120 days, the maximum germination rates recorded are around 96%; 94% and 82% respectively for afares oak, zeen oak and cork oak. **(Fig.5)**

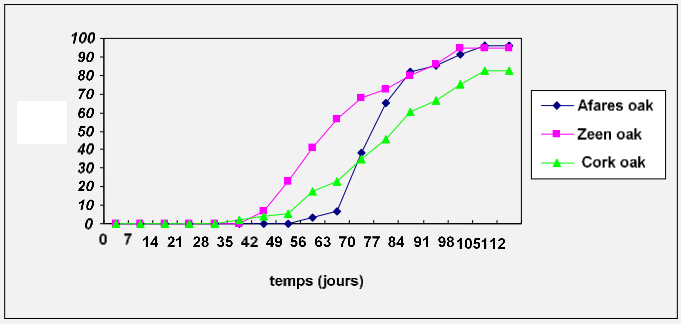

Figure 5: Evolution of the germination rate of acorns of afares oak, zeen oak and cork oak as a function of time (semi-controlled conditions).

3.1. Discussion

Germination is often considered as a phenomenon which leads from inert seed to a seedling capable of emitting its first leaves. This phenomenon presents various modes of expression. The germination rates observed over a period of 120 days were manifested in three phases: firstly a latency phase with a more or less high duration for the afares oak (56d) compared to that of zeen oak (42d) and cork oak (36 days); then a second phase of acceleration of germination (extends from 56 to 105 days for afares oak, 42 to 98 days for zeen oak and from 36 to 105 for cork oak) and a third stage characterized by a tray indicating the maximum rate of germination (96%; 94% and 82% respectively for the afares oak, the zeen oak and the cork oak).

This can be explained by the time required for the seed to set up mechanisms that allow it to adjust its osmotic potential in relation to the medium (Preney, 1997); It should also be noted that there is differential dormancy within the same species or the same batch of seeds. Or even seeds in the same mother tree. The intensity of dormancy varies according to latitude, provenance and year of harvest. This dormancy has the effect of staggering germination over a more or less long period of time (Mokrani,2002).

The germination difficulty by the existence of an embryonic dormancy due to the mechanical action exerted by the pericarp at the exit of the radical and which depends on the producing tree which is one of the characteristics of physiological aspects of ripe and freshly harvested acorns (Merouani,2000). Slow germination is also reported for cereals (Chaussat, 1975) for holm oak (Aissa, 1983). The germination rate decreases when the shelf life increases because it leads to a decrease in the vitality of the acorns (Preney, 1997).

In the nursery, the three oak populations have maximum high germination rates obtained after one month of storage. These results are in agreement with that found by Ncibi, 2005) on the oak species.

Germination also seems to be dependent on the size of acorns. The results which are obtained are in agreement with those found by (Hasnaoui, 1992) who attributes that the size of acorns or, their weights has a positive influence on germination rates and their early stages of growth; The difference in acorn size can be attributed to the ecological and climatic conditions of the origin site's and the specific conditions of the study species.

- The morphological study of the young plants of afares, zeen and cork oaks shows that the heights and the average diameters obtained vary from 4.27; 9.46 and 8.34 cm on March to 9.37; 18.22 and 20.38 cm on July, and 2.36; 3.15 and 2.42 mm on March at 7.95; 6.52 and 5.92 mm on July, respectively in afares oak, zeen oak and cork oak (Fig. 6). Statistical analysis shows a significant difference between the species by the Fisher F test at the 5% threshold.

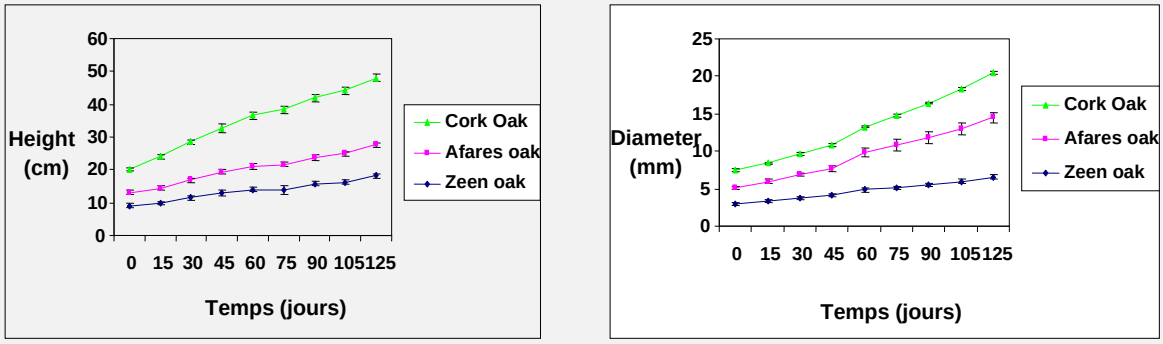

Figure 6: Evolution of the average of heights and diameters of young plants of afares oak, zeen oak and cork oak in the population of Aîn Zana as a function of time.

3.2. Discussion

The growth of the aerial part of young plants of cork oak is greater than zeen oak. On the other hand, it is weaker in afares oak throughout the measurement period (5 months).

Cork oak recorded the highest growth rate (12.04 cm) in stem height and the lowest in diameter at the neck (3.5 mm) despite its small size of acorns (2.93 cm in length and 1.56 cm in diameter). On the other hand, the afares oak presents the lowest growth rate (5.1 cm) in stem height and the highest in diameter at the neck (5.59 mm) with acorns of larger size (4.09 cm in length and 1.72 cm of diameter). This shows that the rate of increase in height and diameter does not depend on the size of acorns but depends on other characteristics specific (physiological, biological) to the species. The oak vegetative cycle is characterized by two periods; a period of active life (in spring and autumn) and a period of slow life (in winter and summer). This manifests an important sensitivity towards low temperatures which are unfavorable for the continuation of its photosynthetic process. This finding is mentioned by (Ehleringer, 1977) and (Acevedo, 1985).

The decline in growth could be due to a decrease in cell extension, which represents one of the first responses to drought (Aloui, 2006).

The morphological characterization of the leaves of young plants shows that the average values of the length and width of the leaves of young plants are 4.43; 7.73 and 4.40 cm and around 2.86; 4.42 and 2.46 cm respectively for the afares oak, the zeen oak and the cork oak (four months old) **(Fig. 7).**

The average lengths of the petioles and the leaf areas are 0.32 cm; 0.52 cm and 0.30 cm and of the order of 8.8 cm^2, 20.9 cm^2 and 7.37 cm^2 respectively for the species of afares oak, zeen oak and cork oak. **(Fig. 8)**

Statistical analysis shows a significant difference between the species by the Fisher F test at the 5% threshold except for the mites and cork.

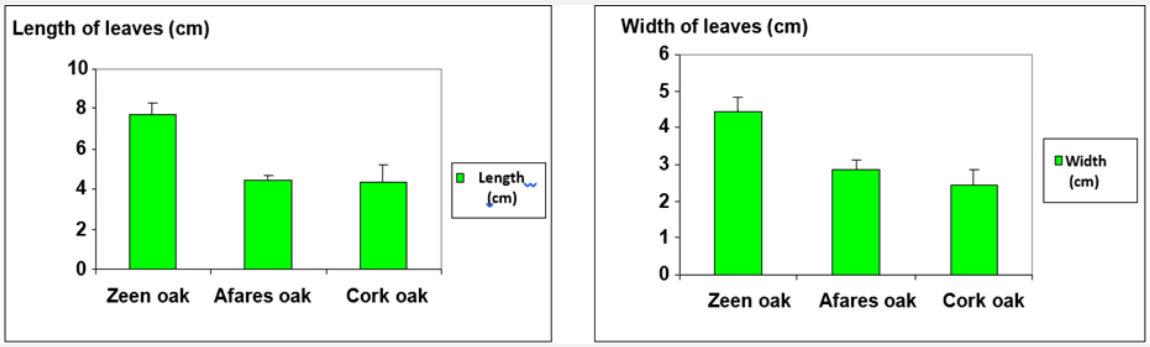

Figure 7: Average lengths and widths of leaves of young plants of afares oak, Zeen oak and Aïn Zana cork oak.

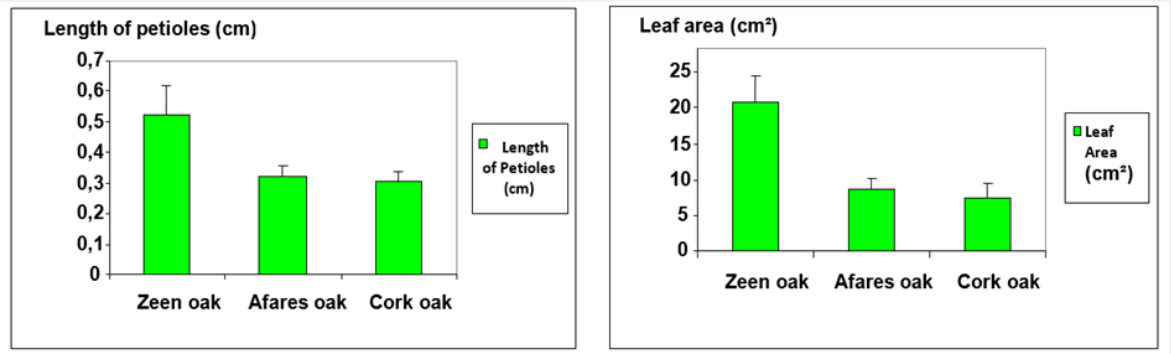

Figure 8: Average lengths of petioles and leaf areas of young plants of afares oak, zeen oak and cork oak in Aïn Zana

Examination of the oak populations studied at the juvenile stage shows that, in addition to variations in leaf dimensions, there is also variation in leaf shapes.
For the afares oak species, the leaves are generally oval in shape, small pennatilobate size with lateral spines; with a slightly wavy edge, those of zeen oak they are generally large, with an oval toothed shape the blade is toothed edge, all these leaves are not very thorny and are all subcessile. Concerning the cork oak, the variability is more important from the point of view of form where we distinguish small and leathery leaves of rounded to sub-rounded shape with small tomentose and whitish teeth below and other microns ovals at the top not very cuneiform at the base lateral ribs are protruding or slightly protruding. All these variations in dimensions and shapes are confirmed by statistical analysis which shows a significant difference between the species of oak by Fisher's F test at the 5% threshold.

The morphological study of the leaves of young plants in the populations studied (afares oak, zeen oak and cork oak) has shown strong morphological variability.
We note that the afares oak leaves occupy an intermediate position closer to the cork oak leaves than those of the zeen oak either for the length, width, length of petioles or for the leaf surface. The juvenile phase is characterized by thornier, shorter-stalked leaves. The leaves of holm oak are larger in size and weight than those of cork oak, as well as the leaves of zeen oak which have greater morphological characteristics than afares and cork oak (Hasnaoui, 1992).
In natural conditions, the morphological study of adult trees of afares oak, cork oak and zeen oak shows that the average values recorded for tree heights and diameters are 16.72, 15.88 and 9.98 m and 0.39 m, 0.25 m and 0.28 m respectively for the species of afares oak, zeen oak and cork oak **(Fig. 9).**

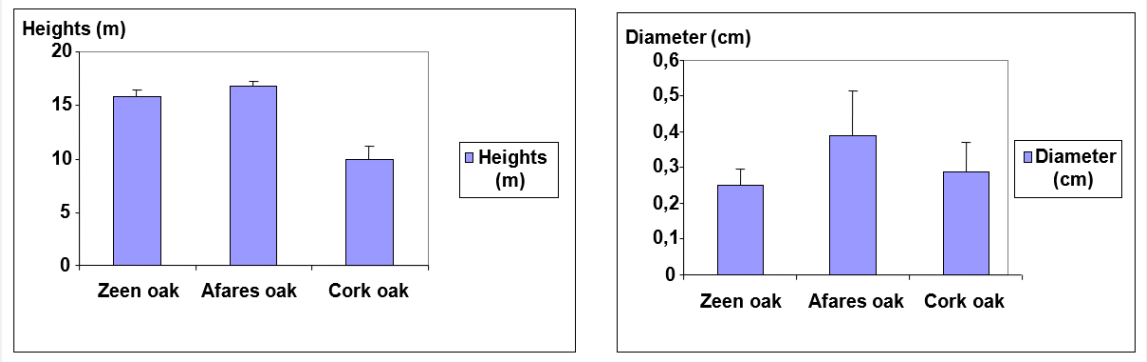

Figure 9: Heights and diameters of adult trees of afares oak, zeen oak and cork oak in Aïn Zana.

The dendrometric measurements carried out on adult trees show that the height is greater in afares oak and zeen oak (around 15 m) than in case of cork oak (around 10 m). This difference in growth could be due to biological factors and genetic factors linked to the species. Thus, physiological activity and the soil type's play an important role by the fact that the trees found on an altitude present the most leached soil, and unlike the age of the trees. This idea is confirmed by (Yessad, 1998) who proves that the height is more important in deep soil than in superficial soil, and in plain than in mountain.
On the other hand, the diameter takes the same values for zeen oak and cork oak (about 0.28 m) and slightly different compared to that of afares oak (about 0.4 m).This may be due to the similarity of

flowering and fruiting in these forest species. The afares oak is a forest species of intermediate form between the zeen oak and the cork oak; this may be due to the mixed stands of this species in the Ain Zana station.

- The morphological characteristics of the leaves of adult trees of afares oak, zeen oak and cork oak shows the following average values of length and width of the leaves of adult trees which are around 11.12 cm, 13.30 cm and 6.93 cm and around 4.11 cm, 8.11 cm and 3.47 cm respectively for the afares oak, the zeen oak and the cork oak (Fig. 10). Statistical analysis shows a highly significant difference between the species by the Fisher F test at the 5% threshold except for the mites and the zeen.

The average petiole length and leaf area are around 1.30 cm, 1.56 cm and 1.29 cm and around 16.25 cm², 40.87 cm² and 14.8 cm² respectively for afares oak, zeen oak and cork oak (Fig. 11). Statistical analysis shows a non-significant difference between the species by the Fisher F test at the 5% threshold.

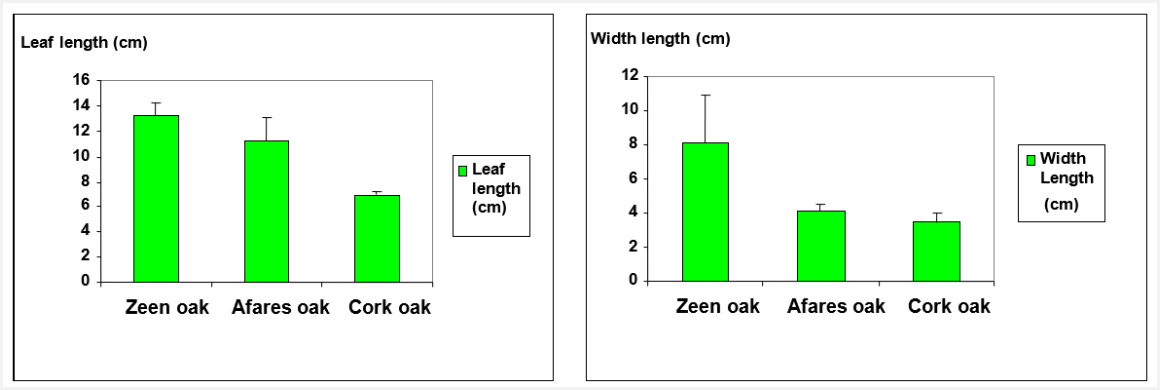

Figure 10: Average leaf lengths and widths of mature trees of afares oak, zeen oak and cork oak in Aïn Zana

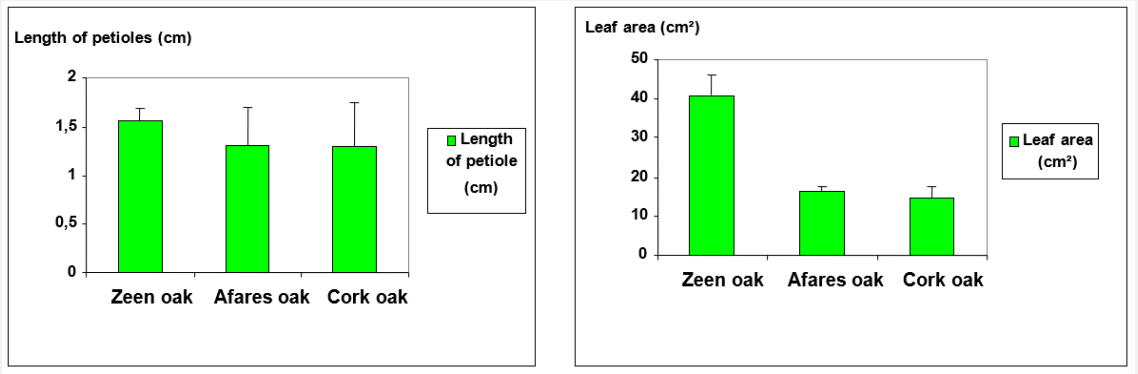

Figure 11: Average lengths of petioles and leaf areas of adult trees of afares oak, zeen oak and cork oak in Aïn Zana.

The morphological study of the leaves of adult trees of the afares oak, zeen oak and cork oak species of the Ain Zana population showed morphological variability, especially between the length of the cork oak species towards the zeen oak and afares oak. The size of the leaves per branch is variable and largely depends on the characteristic of the station and the species (Ncibi, 2005). All this shows that the afares oak has a leaf morphological characteristic identifiable with the zeen oak on the one hand and the cork oak on the other hand which clearly shows their intermediate form between the zeen oak and the cork oak.

4. Conclusion

The present work concerns a comparative study of the morphological behavior, on acorns, young plants and adult trees of afares oak with zeen oak and cork oak carried out in semi-controlled (nursery) and natural (field) conditions.

Germination appears to be dependent on the acorn size factor and the study of oak regeneration by seed has shown that it is possible, but the extent of the latency phase is a limiting factor.

- The growth of the aerial part of young plants is variable depending on the species and the seasons and the increase in height and diameter of afares, zeen and cork oak marks an evolution over time, it is minimal in March and April (Spring). This decrease in growth could be due to a decrease in cell extension which represents one of the first responses to drought.
- The morphological study of the leaves of young plants of the populations of 3 species of oak has shown morphological variability, but we note that the leaves of afares oak occupies an intermediate position closer to the leaves of cork oak than those of zeen oak. The juvenile phase is characterized by thornier, shorter-stalked leaves.

The dendrometric measurements carried out on adult trees show that the difference in growth could be due to biological factors and genetic factors linked to the species with physiological activity resumed in them, to the nature of the soil and to the difference of the age of trees.

- The afares oak is a forest species with an intermediate form between the zeen oak and the cork oak; It is distinguished from other acorn pruning species, germination rate especially with cork oak as well as the increase in height and diameter;

Acknowledgements

This research was supported by grants from the National Institute for Research in Rural Engineering, Water and Forests (University of Tunis Carthage), and the Faculty of Sciences of Tunis (University of Tunis El Manar).

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