RESPONSE OF OLIVE TREE CULTIVARS TO DIFFERENT LEVELS OF NUTRIENT STRESS

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Abstract

To evaluate the response of the olive tree cultivars *Meski* and *Chetoui* to nutrient stress, a complete privation of major nutrients such as nitrogen (N), phosphorus (P), magnesium (Mg), and potassium (K) was chosen. Mineral composition, vegetative development, starch accumulation and their evolutions and distributions in the different compartment of the tree were studied. The survey of mineral diagnosis showed significant modification of the tree mineral alimentation, as well as cationic and anionic at varying degrees. Synergistic and antagonistic actions of the mineral elements were registered, reinforced by the imposed nutrient stress. The distribution of dry matter in the various organs of the olive tree cultivars was affected. The removal of nitrogen seemed to favor the allocation of carbon from leaves to roots. The effect of phosphorus deprivation was marked particularly on the development of the root system which seemed to be small, sparse and poorly developed. Important starch synthesis and storage was also revealed following nitrogen deficiency, but it was very weak after the suppression of magnesium, which may provoke a weakening of growth and development of these young olive trees. Cultivar comparison showed the superiority of *Chetoui* to support the severe imposed nutrient stresses.

Keywords: Olive tree, nutrient stress, mineral diagnosis, vegetative development, starch accumulation

Introduction

Olive (*Olea europaea* L.) has been known as ‘‘the fragrance of the soft gold’’ due to its high economic, social and cultural values. Nowadays, olive is the most extensively cultivated oleiferous tree species in the world, covering an area of 10 million of hectares (Yang et al., 2007). There are more than 1200 olive cultivars. Among them, more than 800 cultivars are for oil production, over 100 are table olives and the rest are used for dual purpose Bartolini et al., 1998). Olive originated in the Mediterranean area, where the production accounts for more than 90% of the world oil production (Wan-Ze et al., 2013). Tunisia is the most famous country for olive culture in the Southern Mediterranean; more than 30% of its arable land is devoted to the cultivation of olives. Tunisia is ranked fourth in terms of number of trees and second in area terms (Karray et al., 2010).

Tunisia, despite its 70 million olive trees, suffers from fluctuations in the annual production which causes adverse consequences in the national economy (GDAP, 2011). As for olive trees, the large charge in flowers or fruits forms a source of competition that emphasizes the production alternation. This alternation depends on the relationship Source-Well. Olive adjusts its charge according to its reserves.

Source-Well relations are closely related to soil fertility. Nerveless, the actual demand for nutrients and detection of mineral deficiencies remain little studied and correction terms of foliar fertilization remain inconclusive.

In a context where fertilization is missing or is practiced empirically in the absence of specific standards, signs of bad nutrition appear and affect productivity. The issue of fertilization is that provide nutrients at the right time and just the right dose to best satisfy the needs of the tree without creating harmful surpluses to the environment.

The diagnosis of the olive grove nutritional status is the only way to determine its nutritional needs at a specific time. Among the methods of diagnosis, which is the more accurate is the foliar analysis. This method is used to identify nutrient imbalances, to assess the level of nutrients before the appearance of deficiencies, to measure the response to fertilization programs and to detect toxicities caused by certain elements.

Thus, the nutritional status of the olive tree is diagnosed after its deprivation for major nutrients such as nitrogen, phosphorus, potassium and magnesium. The impact of these deficiencies is revealed on the growth in dry matter of the olive tree and starch distribution in Well organs.

Material and Methods

Plant Material

Chetoui: This is one of the most important and principal cultivars of oil in Northern Tunisia, where the average annual rainfall is never less than 400mm, it is present in a proportion of 90 to 95%. Its fruits are used mainly for oil extraction, the pulp is easily separated from the core and fruit yield oil is medium (between 19 and 22%).

Meski: is present in North Tunisia, slightly vigorous and rustic. Its productivity is medium and constant. It flowers early, but it is self-incompatible. The most commonly used cultivars as pollinators are *Chetoui*, "*Besbessî*" and "*Picholine Languedoc*". Its fruits are good, they mature early but they have reduced oil contents. This cultivar is resistant to salinity but sensitive to drought and calcareous soils.

Experimentation

Culture was conducted in a greenhouse under hydroponic NFT mode (Nutrient Film Technique), which has the advantage to better control the mineral nutrition of plants, as well as the pH of the root zone, to maintain the availability of trace elements, present in the solution and to eliminate drought problems.

Firstly, young olive plants were transferred from their original substrate to pots, containing a mixture of two inert substrates: perlite and sand, in the proportions of 2/3 and 1/3 respectively. The addition of sand was adopted to increase the capillary, ensure easier ascent of water and ameliorate its retention. While, the presence of perlite increased the porosity and allowed a good ventilation for roots. A control solution (C) was prepared according to Hoagland and Arnon (1938).

This solution had been tested on the olive tree by several authors, such Hatmann and Brown (1965), Braham (1997), Boussadia et al. (2008, 2010) and Saidana et al., (2004, 2006 and 2009) .It contained macro and micro elements necessary for an optimal growth. Four other solutions were prepared by eliminating one major element such as nitrogen (N), phosphorus (P), potassium (K) and magnesium (Mg).

Fertilization took place with these prepared recycled nutritive solutions. For each treatment, 30 plants were tested: 15 for the cultivar *Meski* and 15 for the cultivar *Chetoui* (Figure 1).

Electrical conductivity and pH were continuously controlled and corrected. For all treatments, the conductivity was maintained approximately at 0.92 mS/cm and the pH fluctuated between 5.8 and 6. The correction of the electrical conductivity was done, either by adding the nutrient solution to increase its value, or in some cases, by the addition of a distilled water to decrease it. The pH was checked and corrected by the addition of H_2SO_4 or NaOH

Figure 1. Experimental Station

Mineral Diagnostic

In order to clarify the effect of certain major component deficiencies on the absorption of nutrients and their distribution in the different organs of the olive tree, we followed changes in nitrogen, potassium, magnesium and calcium contents.

To determinate the total nitrogen, Kjeldahl method was used (Martin-Prével et al., 1984).

The total N concentration was calculated according to the following formula: $N\% = n/(10xP)$; where n is the volume in ml of the titrating solution and P is the sample in grams.

The dosage of K, Na and Ca was carried out by flame spectrometer. The spectral emissions of these ions were measured and compared to those obtained from calibration synthetic solutions from 0 to 200 µg K/mL for potassium, 0 to 100 µg Na/mL for sodium and 0 to 10 µg Ca/mL for calcium.

The content of K, Na and Ca in % of dry weight was given by the following formula:

$$\textbf{K, Na, Ca \%} = \mathbf{N \times D \times V / 10^{4} \times P}$$

The dosage of magnesium was carried out by atomic spectrophotometer at a radiation of 285.2 nm. Analysis solutions were diluted in lanthanum chloride which was dissolved in HCl (0.1 N). The optical density was compared to those of the calibration solutions, obtained by mixing a solution of pure Mg at 1 g/L with hydrochloric acid (0.1 N). The standard range of Mg was 0.2, 0.4, 0.6, 0.8 and 1 mg/L.

Evaluation of Growth in Dry Weight

Olive trees were divided into root, stem and leaf fractions. The dry weight (DW) of shoot, leaves and roots was determined after drying at 70 °C for 72 h.

Determination of Starch Content

To evaluate the effect of the different stress levels on the synthesis and the distribution of starch in the olive woods and roots, Nielson’s method was adopted (Braham, 1997). This method is based on the apparition of the blue color following the action of the iodine on the starch which is determined spectrophotometrically.

Statistical Analysis

The data were processed by SPSS 10 in version 2.0, variance and means comparison were analyzed by the SNK method (Student-Newman-Keuls).

Results and Discussion

Effect of The Nutritional Stress on Mineral Absorption of The Olive Tree

Nitrogen content variation

The nitrogen in the control treatment plants seemed slightly deficient compared to the values mentioned by De Monpezat and Denis (1999) and De Monpezat et al. (2000) during the winter dormancy (February), but it was in the standard norms during curing core (June and July). For all treatments, maximum values were recorded in the spring, indicating an important physiological activity, with an increased consumption of nitrogen (Figure 2). These results corroborated those of Garcia et al. (1992), Denis and Afidol (2000), and Bustan et al. (2013) who reported an increase in nitrogen consumption via two peaks of high demand, the first in starting vegetation and the second in flowering stage.

The varietal comparison revealed that nitrogen levels were significantly higher in *Chetoui* than in *Meski* for all treatments. In addition, the statistical analysis showed a significant difference in the nitrogen contents of plants deprived for nitrogen and witness, in the both cultivars *Chetoui* and *Meski*. A reduction of 46.64 and 42.67 % was mentioned respectively.

The imbalances in the levels of P, K and Mg seemed to have bad effects on the nitrogen absorption. Indeed, there was a decrease in the nitrogen content of 6.15, 4.59, 11.46 and 9.00, 9.55, 14.87%, following the privation of P, K and Mg, respectively in *Meski* and *Chetoui*. Therefore, the removal of magnesium from the nutrient solution had the most serious effect on the absorption of nitrogen. The effect of the phosphorus privation on nitrogen absorption was confirmed by Hartmann and Brown (1953), indicating that a marked deficiency in P leaded to an abnormally low level of N, while P fertilization increased this level.

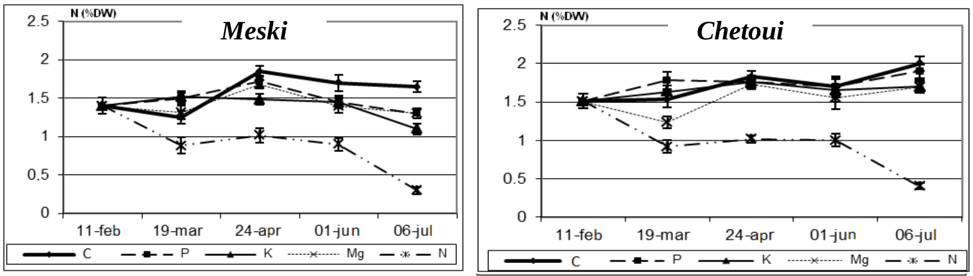

Figure 2. Evolution of total nitrogen concentration of *Meski* and *Chetoui* cultivars under five nutritional levels (C, P, K, Mg and N)

Nitrogen content distribution

The accumulation and the distribution of nitrogen shown in figure 3 seemed to be strongly affected by the different applied deficiency levels. After three months of experimentation, a strong accumulation of nitrogen was noted in roots of the both experienced cultivars deprived for nitrogen. The nitrogen accumulation in the roots was approached to that of the leaves, which resulted a significant increase of dry weight of the underground part in order to the aerial part one. However, at the end of the test, the distribution of nitrogen into the underground part decreased and vanished; root growth seemed to be probably strongly affected.

Similarly, the potassium privation in the nutrient solution limited the nitrogen level in the treated plants. This effect increased over time, and was particularly noticeable in the underground part. In fact, a nitrogen content reduction of 23 and 16% was observed in the roots of both cultivars *Meski* and *Chetoui*, respectively after three months of testing. This reduction increased and the nitrogen content vanished in the roots at the end of the test. This result confirmed the low root development in plants deprived of potassium. The test duration appeared sufficient to detect the potassium privation effect on the development of the cultivar *Meski* roots. It is important to keep this test trying to see the result of this effect on the cultivar *Chetoui*.

Following the phosphorus privation treatment, the cultivar *Meski* appeared more sensitive, with nitrogen-free root content. Unlikely, the cultivar *Chetoui* showed a decrease of 27.77%, compared to the control, at the end of the test.

The effect of magnesium deprivation seemed slightly marked on the nitrogen uptake comparatively to the effect of the other nutritional levels. In fact, we noted a decrease of 7.6% in the nitrogen content of *Chetoui* roots and 15% in that of *Meski* woods.

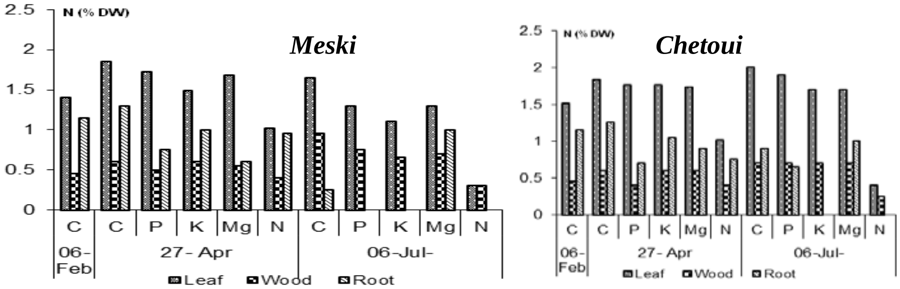

Figure 3. Evolution of total nitrogen distribution in the different organs (leaves, woods, roots) of *Meski* and *Chetoui* cultivars under five nutritional levels (C, P, K, Mg and N)

Potassium content variation

The variations in potassium concentrations under five nutritional levels, for the both cultivars *Meski* and *Chetoui*, were presented in figure 4. In both cultivars, the foliar potassium content in the treatment (K) was absolutely the lowest. Indeed, the leaf content of this element fell significantly as 20.25% for *Meski* and 19.43% for *Chetoui*, comparatively to the control.

The highest foliar potassium content was recorded for the treatment (Mg), both in *Meski* and *Chetoui*. This suggested an antagonism for these two elements, in accordance data reported by Gonzalez and Troncoso (1972). Despite the loss of phosphorus, *Meski* and *Chetoui* presented foliar potassium contents similar to those of the witness.

Once more, it is important to note, in April, an increase of foliar potassium content was similarly noted for all applied treatments on the both cultivars. Boulouha and Elboustani (1995) explained that in such time, the potassium accumulation tendency was to avoid the metabolism orientation towards the synthesis of amino acids which are not favorable to the floral induction.

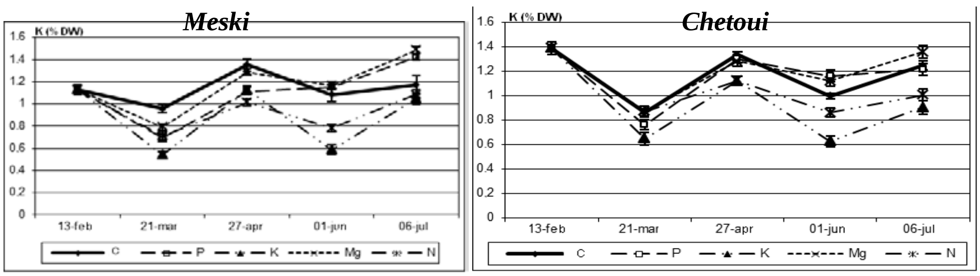

Figure 4. Evolution of potassium concentration of *Meski* and *Chetoui* cultivars under five nutritional levels (C, P, K, Mg and N)

Potassium content distribution

The evolution of K distribution in the leaves, woods and roots, represented in the figure 5, showed that, in the case of the treatment (K), the decrease in the accumulation of the potassium, was widespread in the different parts. This relative decrease was more pronounced in roots than in woods. The allocation of this major element to the roots appeared limited by the nutritional stress. Indeed, there was a decrease of 27.65 and 40.4% respectively in the root growth of the cultivars *Meski* and *Chetoui*. While, decreases of 12.76 and 24.75% were unregistered respectively in the wood dry weights of these cultivars. Consequently, after the leaves, the accumulation of K seemed to be principally in woods.

After treatment (Mg) and contrary to what is obtained for (K), we noted an increase of the potassium accumulation in all organs of both cultivars. This increase was 17.14, 23.68, 21.05 and 28.75, 10.25, 8.04% respectively in the roots, the woods and the leaves of *Meski* and *Chetoui.* For this treatment, the potassium accumulation appeared principally to occur in the woods of the cultivar *Meski* and in the roots of the cultivar *Chetoui.*

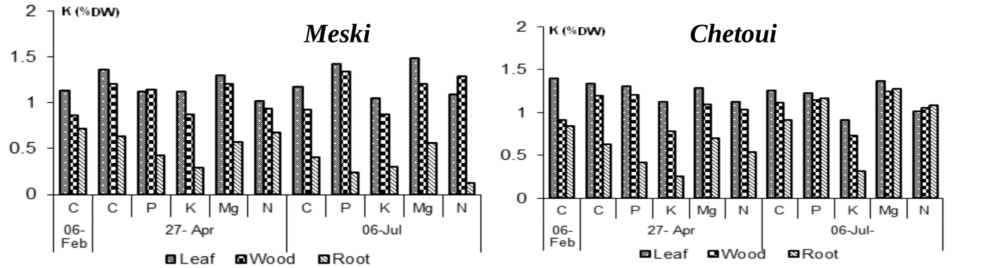

Figure 5. Evolution of potassium distribution in the different organs (leaves, woods, roots) of *Meski* and *Chetoui* cultivars under five nutritional levels (C, P, K, Mg and N)

Magnesium content variation

The evolution of the magnesium foliar content, shown in figure 6, seemed affected by the nutrient imbalance applications. For both cultivars, the foliar magnesium content in the control treatment remained in standard values during winter dormancy (0.12 to 0.14% in February) and during the pit hardening (0.08 to 0.10 in June and July). The lowest magnesium foliar content was recorded following the magnesium privation treatment, with decreases of 12.68 and 6.7% respectively for *Meski* and *Chetoui.*

Potassium deficiency appeared to induce an excess of magnesium content, illustrated by an increase of 3% in the cultivar *Meski* and 3.67% in the *Chetoui*. This result seemed to confirm that of Gonzalez Troncoso (1972).

A slight increase in the magnesium content was noted following the phosphorus private treatment in both tested cultivars, which seemed to differ from the results discussed by Brown and Hartmann (1953), according to them, a marked deficiency in P leaded to abnormally low levels of Mg.

Similarly, the nitrogen deprivation seemed to increase the foliar magnesium content. According to Garcia et al. (1992), the levels of magnesium were associated with soil fertility, while climatic factors had not effect.

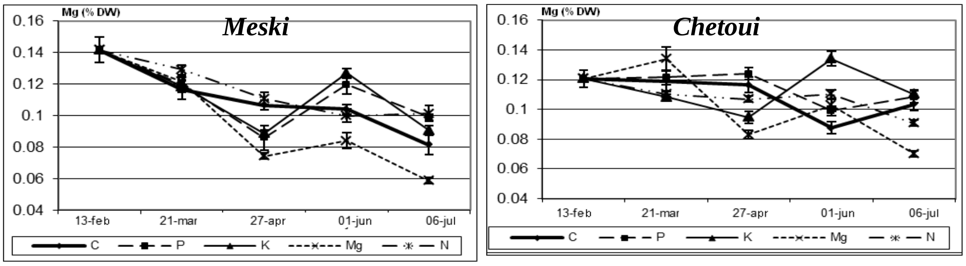

Figure 6. Evolution of magnesium concentration of Meski and Chetoui cultivars under five nutritional levels (C, P, K, Mg and N)

Magnesium content distribution

The accumulation and the distribution of magnesium in the different organs of both tested cultivars *Meski* and *Chetoui* were shown in figure 7.

In response to the magnesium privation, it appeared that the leaves were the first affected, the magnesium content reduction was then generalized in the different organs of the olive trees, it was respectively 27.84, 46.59 and 10.37 in the leaves, the woods and the roots of the cultivar *Meski* and 37.03, 17.64 and 28.65 in those of the *Chetoui*.

Nutrient imbalances in potassium and in nitrogen exerted early effects on the magnesium contents. Concerning the whole plant, increases of 12.00, 22.89 and 13, 18.34% were already registered, respectively in both cultivars *Meski* and *Chetoui* in the first months of the experimentation, which demonstrated the existence of an antagonism between N, K and Mg and synergy between N and K.

Following the application of the treatment (P) and concerning the whole plant, an increase in Mg contents was recorded in both cultivars, but at the end of the test and in the cultivar *Meski* a decrease of Mg concentrations was noticed in the roots. It will probably widespread in the rest of the organs if the test continues, which may confirm the results described by Hartmann and Brown (1953).

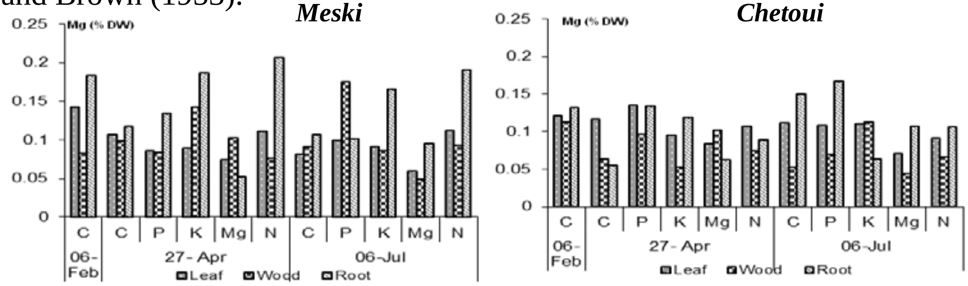

Figure 7. Evolution of magnesium distribution in the different organs (leaves, woods, roots) of Meski and Chetoui cultivars under five nutritional levels (C, P, K, Mg and N)

Calcium content variation

Calcium is a structuring element of plant tissue, firstly by its binding power and its capacity for coordination between macromolecules and secondly with the stability and reversibility of its bonds at the cell membrane (Marschner, 1995). According to Huglin (1986), it has the highest levels comparatively to the other cations, with low absorption kinetics compared to the potassium (Mengel and Kirkby, 1987).

Under the five levels of the nutritional stress, changes in foliar calcium in the both studied cultivars were represented in the figure 8.

For both cultivars, the foliar calcium contents for the control treatment remained in standard values, between 1.75 and 2.25%, and the same for the woods. Differently, calcium levels were higher in the roots. Their values were 1.94% for the cultivar *Meski* and 1.36 % for the *Chetoui*, while the standard values varied in the range of 0.8 to 1.0%. In this fact, despite the importance of the calcium root uptake, both cultivars seemed to settle their calcium content in both leaves and woods.

Varietal comparison showed no significant differences in the concentrations of calcium, but the comparison inter-treatments led to a significant effect.

Statistical analysis revealed a significant difference between the control and treatment (Mg) for the cultivar *Meski*, this result confirmed the existence of a competition between calcium and magnesium.

Concerning the cultivar *Chetoui*, there was a significant difference between calcium contents in the treated olive trees private (K) and those private (Mg).

Mg and K have an interaction with calcium. Indeed, in the absence of potassium, calcium became lower in olive trees private (K) than in control. In contrast, the removal of magnesium in the nutrient solution seemed to increase the foliar calcium content.

The interaction between the elements, calcium, potassium and magnesium had been reported by several authors, Hartmann and Brown (1956), Gonzalez and Troncoso (1972), Garcia et al. (1999), according to whom, calcium absorption is influenced by the presence of other cations, essentially magnesium and potassium which can create significant competition according to the plant request.In the case of the treatment (N), foliar calcium content seemed to be low, in the order of 0.93% for *Meski* and 1.01% for *Chetoui*. This result leaded us to assume the existence of a synergy between nitrogen and calcium.

Kirkby and Knight (1977) and Graham (2001) had confirmed it. Indeed, according to Coïc (1963) and Graham (2001), this correlation was related to the form assimilable of nitrogen in the soil solution or nutrient solutio (hydroponics).In the case of treatment (P), the calcium foliar content of both cultivars *Meski* and *Chetoui* took high values, but not significant comparatively to the control at the level of 5%. Contrarily, Gavalas (1973) affirmed that a marked P deficiency leads to abnormally low levels of calcium.

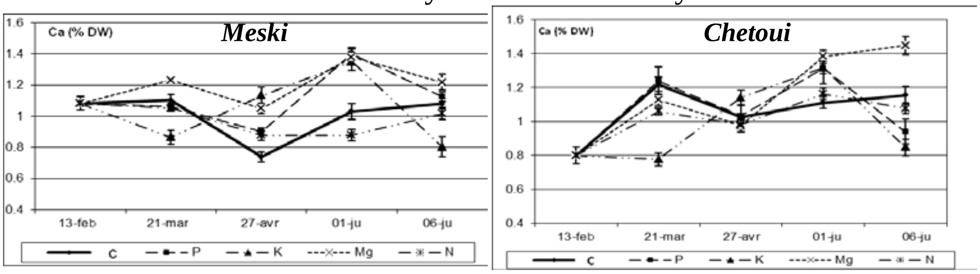

Figure 8. Evolution of calcium concentration of Meski and Chetoui cultivars under five nutritional levels (C, P, K, Mg and N)

Calcium content distribution

Shown in the figure 9, the distribution of calcium in the various organs of both cultivars *Meski* and *Chetoui* appeared to be strongly modified by the nutritional stress. In this context, there was an increase of 13.14 % in the calcium levels in the wood of the cultivar *Meski* for (Mg) treatment, comparatively to the control, and a decrease of 24.49% in its roots. While, an increase of 63.46%, in the calcium rate of the cultivar *Chetoui* roots was recorded, and only 39% in the wood of this cultivar comparatively to the control. The accumulation of Ca seemed to be mainly in the roots of the cultivar *Chetoui* and in the woods of the cultivar *Meski*. This result was similar to that obtained in treatment (P). In this fact and comparatively to the control, there was an increase of 29.35% in the calcium rate, in the woods of the cultivar *Meski* and a decrease of 7.84 % in its roots, and an increase of 34.22% in the calcium rate in the wood of the cultivar *Chetoui* and 50.63% in its roots.

For treatment (K), the accumulation of Ca seemed to be mainly in the roots for both studied cultivars.

Concerning the treatment (N), there was a significant decrease in calcium levels either in woods or roots of the cultivars *Meski* and *Chetoui*. The result seemed to be consistent with that found in peach by Graham (2001). This author noted that the levels of Ca in leaves, roots and woods were all significantly decreased according to the decrease of the ratio NO_3-N / $NH4^+$-N (percentage of total nitrogen) in the nutrient solution.

From all these observations we conclude that the mode of distribution of Ca varied depending on the cultivar and treatment. The statistical analysis of cultivar and treatment effects was highly significant at 5%. Similarly, the interaction between these two factors was highly significant at this level.

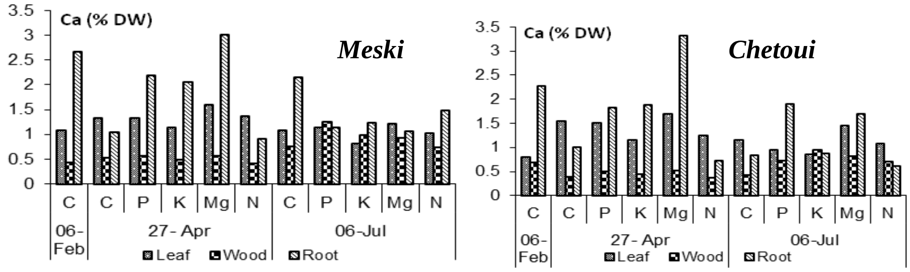

Figure 9. Evolution of calcium distribution in the different organs (leaves, woods, roots) of Meski and Chetoui cultivars under five nutritional levels (C, P, K, Mg and N)

Sodium content variation

The variation in leaf sodium content in different types of nutrient stress was shown in the figure 10, respectively for the two cultivars *Meski* and *Chetoui.* The representative curves showed values higher than those reported by Loussert and Brousse (1987). According to these authors the sodium content varied in the range of 0.012 to 0.28%, while the values recorded for the both studied cultivars exceeded this limit and varied from 0.18 to 0.39%. This result can probably be explained by a rise in the concentration of the nutrient solution, in view of the intense evaporation, hardly controlled caused by the high temperature in the greenhouse culture at that time.

The statistical study does not provide a significant difference between treatments and even for treatment (K), the result seemed different from what is found on the vine "Black Grenache" by Ezzilli (1996), who reported that in Tunisia, in a dry year, an accumulation of sodium in the leaves of this plant accompanied the decrease in potassium. Olive tree, whether it is the cultivar *Meski* or *Chetoui*, avoided in this case, leaf sodium accumulation caused by a decrease in the rate of potassium and then seemed to be more tolerant to the stress.

Recall that sodium is an element to which plants show only a very little need; indeed it can enter in cells, which however tend generally to repress it.
Several crops tolerate well the sodium, either because of its low concentration in the phloem, case of cereals (Zid 1983), or by effective vacuolar compartmentalization.

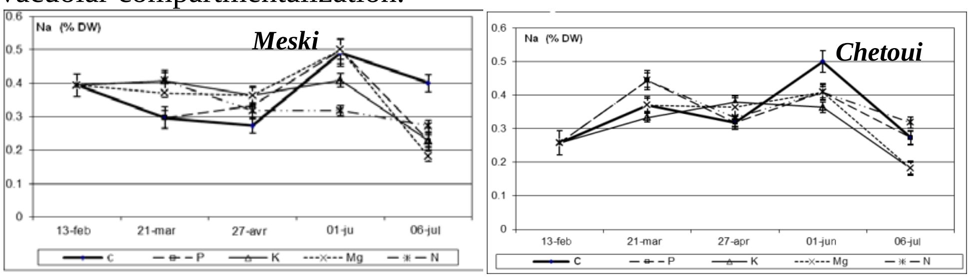

Figure 10. Evolution of sodium concentration of Meski and Chetoui cultivars under five nutritional levels (C, P, K, Mg and N)

Sodium content distribution

The distribution of sodium, shown in the figure 11, indicated that the olive tree seemed to be resistant to salinity despite deprivation of the main major elements such as nitrogen, potassium, magnesium and phosphorus. The sodium foliar content in treated plants was practically the same as that of control treatment.

Sodium levels in the leaves, woods and roots followed an increasing gradient. So, this species seemed arrive to minimize sodium accumulation in leaves and concentrate it in its underground parts.

We note, in the control treatment of the cultivar *Meski*, a sodium accumulation of 0.55% was noted in the roots, 0.39% in the woods and 0.38 % in the leaves. For *Chetoui* this accumulation was 0.63% in the roots, 0.35% in the woods and 0.28% in the leaves. Then, we deduce that the cultivar *Chetoui* appeared more resistant to salinity than *Meski* by minimizing the maximum leaf sodium accumulation.

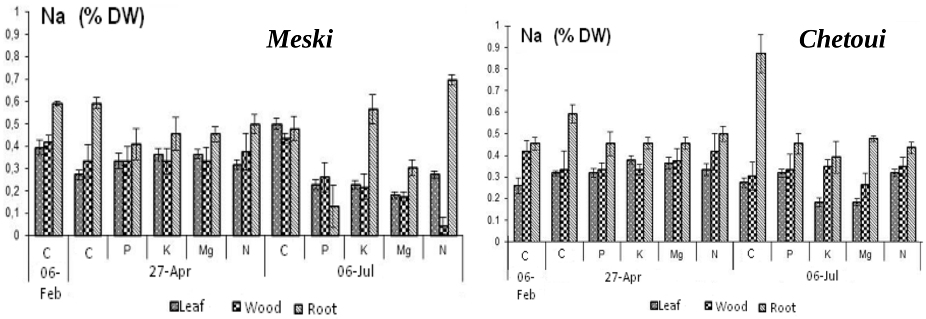

Figure 11. Evolution of sodium distribution in the different organs (leaves, woods, roots) of Meski and Chetoui cultivars under five nutritional levels (C, P, K, Mg and N)

The Nutritional Stress Effect on The Dry Weight Accumulation and Distribution

The nutritional stress effect on the growth in dry weight

The nutritional stress effect on the accumulation of total dry weight (DW) was shown in the following tables 1 and 2.

The control plant of the cultivar *Meski* showed an important increase in DW after the first 3 months, reaching 47.71%, while the DW of the plants subjected to treatment (P), (K) and (Mg) were respectively 8.82, 29.11 and 25.74%, representing thus 18.48, 61.02 and 53.94% comparatively to the control.

Consequently, plants deprived of phosphorus seemed to present the minimal growth. *Meski* cultivar appeared to be most sensitive to the lack of

P, during 80 days of experimentation; then its sensitivity decreased with treatment (Mg) and (K).

Similarly, in the absence of nitrogen, the total DW was strongly affected, then there was a complete inhibition of growth, the plants have a low DW representing only 34.52% of that of the control treatment; so, the effect of the nitrogen lack was both early and also significant at 5%. This result was consistent with what had been widely discussed in the literature: Ramalho et al., (1999) and Vos et al., (2005) suggested that the photosynthetic capacity of leaves was strongly dependent on the nitrogen nutrition under strong illumination.

A good nitrogen supply promoted the functioning of photosynthesis and therefore plant growth, while nitrogen privation reduced the number and size of leaves, shortened the growing period and slowed down the transmission rate and leaf expansion.

The comparison between treatments, along 5th month of experimentation, revealed a significant difference between plants receiving a complete nutrient solution and plants deprived of magnesium. The total dry weight of plants deficient in magnesium represented only 25.19% of that of the control. Indeed, the effect of magnesium deprivation was late and became significant at the fifth month of the experimental period.

During the first three months, the total dry weight of *Chetoui* plants increased by 24.46%, comparatively to the control treatment. While, DW of plants deprived (P), (K), (Mg) or (N) were respectively 6.32, 27.61, 39.58 and 8.29%. Nitrogen and phosphorus seemed to be the both main factors that can greatly affect the general growth of the treated plants, which confirmed their essential role in the growth and the development of plants. The reduction of the plant growth attended, in the treatment (P), 74.16%, and in the treatment (N) 66.10%, comparatively to the control treatment. At the end of the test, this reduction became only 34.56% for the first treatment and 43.56% for the second.

After fifth month of (Mg) privation, the cultivar *Chetoui* already succeeded to tolerate the imposed stress. Indeed, no significant differences in dry weight comparatively to the witness was noticed for this treatment but the effect was only limited to a presence of slowed growth.

Comparing at the end of the test the growth in DW of both studied cultivars with the witnesses, we can suggest for both a sensitivity grid against these nutritional stress levels and in descending order respectively: the absence of magnesium and nitrogen, phosphorus and finally potassium for the cultivar *Meski* and nitrogen, phosphorus, potassium and magnesium for the cultivar *Chetoui*. Therefore, the varietal difference is highly significant (at 5%).

Table1. Average of *Meski* cultivar dry weight in gram

	C	P	K	Mg	N
February	45.03	45.03	45.03	45.03	45.03
April	86.13	49.39	63.53	60.64	29.73
July	144.10	85.10	73.10	36.30	45.00

Table 2. Average of *Chetoui* cultivar dry weight in gram

	C	P	K	Mg	N
February	27.16	27.16	27.16	27.16	27.16
April	35.96	29.00	37.53	44.96	29.62
July	103.00	67.40	115.70	88.60	58.00

The nutritional stress effect on the dry weight distribution

The evolution of the dry weight distribution in the different parts of the tree, according to the applied treatment, was represented by the figure 12. It showed that for the cultivar *Meski* and in the absence of nitrogen, the distribution of dry weight to the root part became increasingly important. At the end of the test, the level of root growth reached 21.92%, which represented an increase of 16.65% more than that in the control treatment.

This result had been confirmed by several authors such as Michaud and Yelle (1994), and Limami Ameziane (1997). These researchers reported that a limited supply of nitrogen in the culture medium promoted allocation of carbon from leaves to roots.

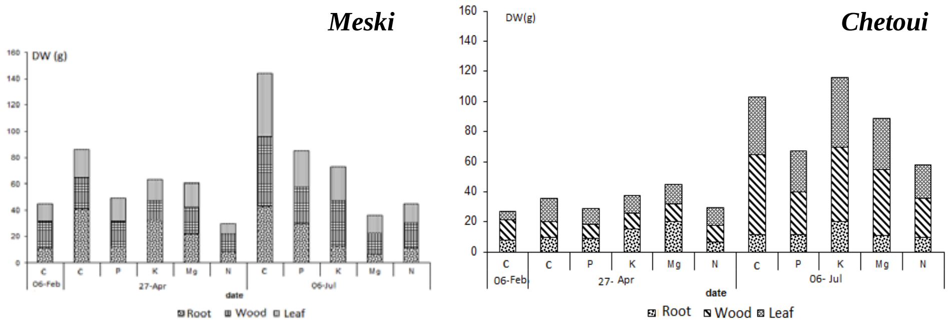

Figure 12. Evolution of the dry weight distribution in the different organs (leaves, woods, roots) of Meski and Chetoui cultivars under five nutritional levels (C, P, K, Mg and N)

Concerning the treatment (P) and in the first months of the experiment, a highly significant decrease of root growth was recorded for the cultivar *Meski*, since the dry weight of the underground part was only 29.45% comparatively to the control. Growth represented then only 10.67% compared to that of the control. At the end of this test, root dry weights increased 39.84%. It is then found that phosphorus is a growth factor that acts specifically on the development of the root system, its action is especially important in young stages.

For the cultivar *Chetoui*, at the end of the experiment and in the absence of nitrogen, the root growth was 45.39%, its dry weight was equal to 89.60% of that of the root part of the witness. While the aerial part dry weight was only 53% compared to the control. The root part was then the largest and the most developed.

Unlike the cultivar *Meski* and under the nutritional level (P), the cultivar *Chetoui* presented a significant root growth, almost similar to that of the control, which leaded to assume that at the root level, the cultivar *Chetoui* remained the most tolerant to a deprivation of phosphorus.

The absence of magnesium in the nutrient solution for 6 months had limited significantly the growth in dry weight. This reduction was generalized in all *Meski* organs, a decrease of 85.58% was noted in the leaves, 69.18% in the woods and 71.36% in the roots, comparatively to the control. This result differed from that found in *Chetoui*, where the lack of magnesium does not mark any significant difference with the control at the level of 5%.

Following the lack of potassium in the nutrient solution, a growth reduction of 49.27% was recorded in the treated plants of the cultivar *Meski*, comparatively to the control. This reduction was 70.23% in the roots, 34.97% in the woods and 46.26% in the leaves. So, the roots seemed to be the most affected by the absence of potassium.

This result confirmed the beneficial role of potassium in the resistance to water stress. Indeed in the absence of potassium, the root system is poorly developed, which limits its ability to absorb the maximum amount of water required to satisfy the needs of the tree. Contrarily, the cultivar *Chetoui* showed a good resistance to stress during the testing period.

The general aspect of *Meski* and *Chetoui* cultivars was described following the different nutritional levels in the illustrations 1 and 2.

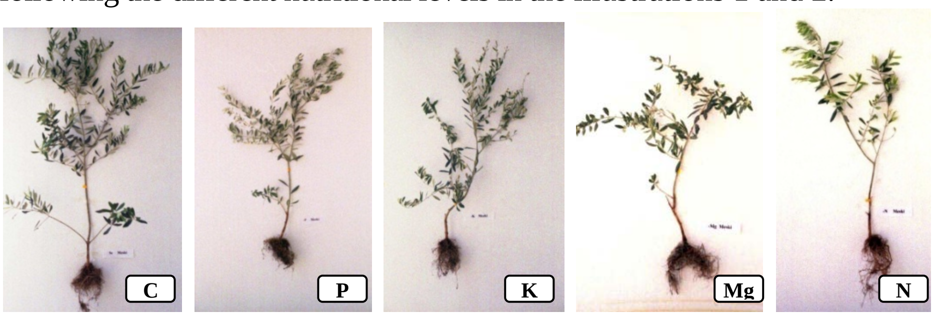

Illustration 1. General aspect of *Meski* cultivar under five nutritional levels (C, P, K, Mg and N)

C: significant growth of the trunk, which presents several shoots.

P: small plant showing a significant drop in terminal leaves.

K: languid vegetation with stunted appearance leaves, especially those of the extremities and therefore a plant sparse top.

Mg: general reduction in plant growth, followed by a significant fall and death of young shoots (the fourth shoot)

N: significant reduction in growth, with little ramification and marked defoliation mostly of the older leaves (the second branch in the right).

Illustration 2. General aspect of *Chetoui* cultivar under five nutritional levels (C, P, K, Mg and N)

C: important extension of the trunk. Shoots well developed.

P: no trunk extension with significant fall leaves

K: Reduce trunk growth. Leaves with vertical position, to minimize luminance reception and escape the desiccation imposed by the absence of potassium

Mg: trunk elongation reduction. Developed foliage but with a sickly appearance

N: no trunk development and a sharp reduction in overall plant growth. Dull and sparse foliage caused by a significant leaves fall, especially the oldest one.

To better illustrate the effect of nutritional stress on the distribution of dry weight in the different plant organs, the distribution ratio (DR) was determined.This was the ratio of the dry weight of the aerial part (leaves and woods) and the dry weight of the underground part. The results, shown in the figure 13, indicated that this ratio was strongly affected by the nutritional stress which acted differently depending on the concerned cultivar. The cultivar *Chetoui* had the higher ratio. This was due to the low dry weight accumulation in the root part of the cultivar *Chetoui*.

The DR of the *Meski* control seemed to be low during the experimental period, reflecting the importance of the root part in relation to the aerial one. Contrary to this report, the DR of the treatment (P) was important during the first three months, the translocation of dry weight from the aerial part to the roots, steeled low.

For other nutritional levels, a considerable increase of 23.01, 51.57 and 50.09% was recorded for the treatments (N), (Mg) and (- K), comparatively to the control. The underground part of plants deprived for nitrogen was then the largest, comparatively to that of the other treatments, by presenting the lowest ratio.

The cultivar 'Chetoui" presented similar distribution ratios for treatment (K), (P), (Mg) and (N), their aerial parts were less developed than those of the control treatment plants.

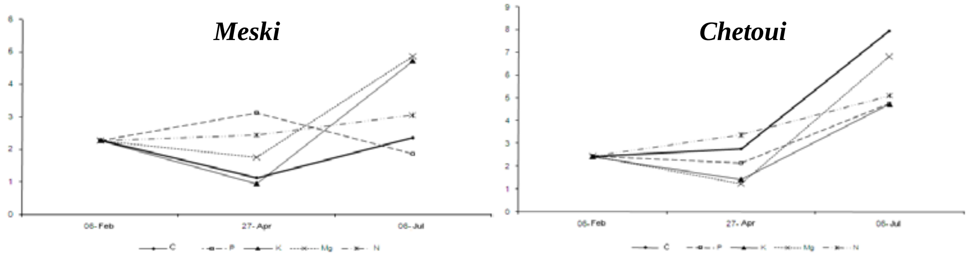

Figure 13. Evolution of the ratio of aerial and underground parts dry weight of Meski and Chetoui cultivars under five nutritional levels (C, P, K, Mg and N)

The Nutritional Stress Effect on the Starch Accumulation and Distribution

The starch rate and its distribution in the woods and the roots under different levels of nutrient stress were shown in the figure 14.

The treatment which had the greatest effect and modified greatly the starch content was the nitrogen privation treatment. This effect was relatively similar in the two studied cultivars, but with a different distribution. Indeed, in the woods, the increased accumulation of starch in plants of both cultivars *Meski* and *Chetoui* was 3.9 times that of the control, however, in the roots, this increase was only 41, 66% in the cultivar *Meski* and 83.2% in the *Chetoui*.

The translocation of photo-assimilates to the roots seemed to be much higher in the cultivar *Chetoui* than *Meski*. Varietal difference was highly significant at 5%. Nitrogen occurs a decisive role in the carbon assimilation and distribution of photo-assimilates between aerial and underground organs, which were widely discussed in the literature.

Limami and Ameziane (1997) reported that the allocation of photo-assimilates to roots increased when the availability of nitrate in the medium decreases.

Rufty et al. (1988) reported that nitrogen starvation also increased the carbohydrate content of the tissues especially in the roots.

By analyzing the results related to treatment (P), two observations could be identified:The rate of starch was higher in the cultivar *Chetoui* than in *Meski*. An increase of 38.36 % in the woods and 4.73% in the roots of the

Chetoui were noted, comparatively to the control. However, in the *Meski* the accumulation of starch was only 97.83% in the woods and 76.89% in the roots.The distribution of starch between aerial and underground parts seemed to be considerably in the woods than in the roots of both studied cultivars.

The treatment (K) didn't affect the rate of the starch in the woods of the cultivar *Meski* in a significant way. This concentration was 94.37% comparatively to the control. In contrast, in roots, there has been a significant decrease of 29% comparatively to the control. This reduction can be explained by a gradual slowing of the translocation of assimilates fault of the absence of potassium.

This fact corroborates the observations of Michaud and Yelle (1994), whose confirmed the existence of a significant influence of the potassium on the source-sink relationships. Indeed, in the presence of a nutritional deficiency, there is increasing amounts of abscisic acid which in turn exert a major influence during the unloading of assimilates. Potassium deficiency causes a migration and a storage inhibition of carbohydrates in storage organs.

The results, after treatment (K) for the *Chetoui*, seemed consistent with those found for *Meski* but with higher concentrations. In fact, in the *Chetoui* woods, there was an increase of 14.33% comparatively to the control. However, the rate of starch in the roots was only 95.44% of that of the control treatment.

Magnesium deficiency affected greatly the rate of starch. Indeed, receiving for six-months a nutrient solution deprived of magnesium, the treated plants of both cultivars *Meski* and *Chetoui* could no longer synthesize carbohydrates. Starch concentration vanished at the end of the test and starch was not stored. This leads us to deduce the importance of magnesium in the synthesis and the reservation of carbohydrates and confirm what is reported about this in the literature.

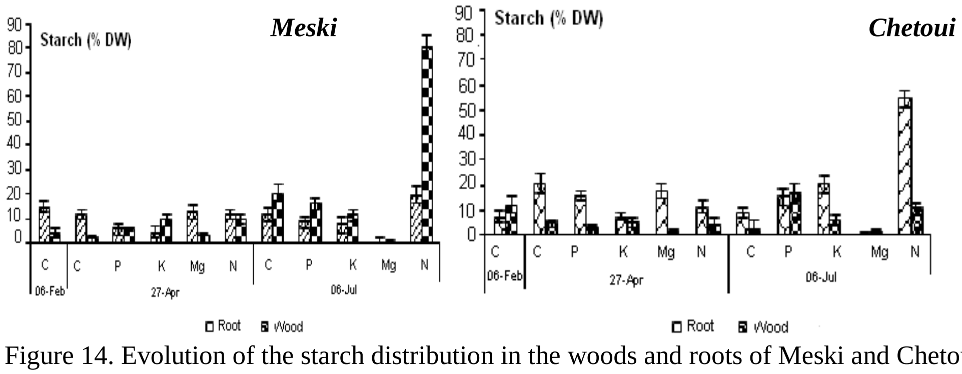

Figure 14. Evolution of the starch distribution in the woods and roots of Meski and Chetoui cultivars under five nutritional levels (C, P, K, Mg and N)

Conclusion

The cultivar *Meski* seemed to be more sensitive to nitrogen deficiency and had the minimum dry weight growth. Such sensitivity was comparable to that caused by magnesium deprivation, but seemed to decrease respectively with the phosphorus and the potassium.

It is important to note that the effects of the lack of nitrogen and phosphorus on the growth in dry weight seemed to be early, while those due to the loss of magnesium and potassium seemed to be late.

Deprivations of P, N, Mg and K have the same effect on both cultivars but with a time lag for the cultivar *Chetoui*.

To better identify the organ, which showed clearly the effect of the imposed nutritional losses, the distribution of dry weight in the different organs of the olive tree was studied. Removing nitrogen seemed to favor the allocation of carbon from leaves to roots, reflected by a low report of the dry weight of the aerial part /underground part. The effect of deprivation of phosphorus was showed particularly on the development of the roots part but unlike private nitrogen, this part showed a reduced sparse and was poorly developed.

The mineral diagnostic study, during the nutritional deficit, showed a huge modification of the mineral nutrition of the olive tree, both the cationic and the anionic one with variation degrees for the two studied cultivars. The obtained results allow us to highlight some heterogeneity in the distribution of major mineral elements within compartments of the tree and infer synergistic and antagonistic actions reinforced by the imposed nutritional stress.

Deprivation of phosphorus and magnesium conducted to abnormally low levels of nitrogen. On the other hand, the lowest potassium levels were recorded for the privative nitrogen treatment. Deficiencies in nitrogen and potassium appeared to cause synergistically the installation of a water deficit. The highest potassium levels were obtained in contrast after treatment without magnesium, showing the antagonism between the actions of these two elements.

The calcium absorption appeared to interact with the contents of potassium, magnesium, phosphorus and nitrogen of the treated plants. In the absence of potassium or nitrogen in the nutrient solution, the rate of calcium decreased relatively to the control, while it increased following the removal of magnesium and phosphorus. Such modifications of macro element contents, as well as their distribution in the different organs of the experienced plants underlined structural and chemical changes that may have more or less a direct impact on the physiology of the plant.

To better understand the effect of nutritional stresses on the metabolism of the two tested cultivars, a study of the starch rate evolution was conducted according to the different nutritional levels. Nutritional stress seemed to have a great influence on the source-well translocation. Starch contents showed an increase, whose importance varied according to the considered treatment. So, due to the lack of nitrogen in the nutrient solution, starch contents were intensely high, indicating an important adaptation mechanism and a better reservation against the stress.

This result is very interesting because it can provide explanations for the high proportion of the dry weight produced in the roots of stressed plants. Magnesium deficiency had greatly affected the levels of starch, it completely inhibited its synthesis and its reserve and thus caused a significant weakening of the growth and the development of the deficient plants.

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