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EFFECT OF MINERAL DEFICIENCY ON MORPHOLOGICAL AND PHYSIOLOGICAL PARAMETERS AND ON SHOOT AND ROOT MINERAL PARTITIONING IN THREE SUNFLOWER VARIETIES

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SUMMARY

An experiment has been carried out in order to study the behaviour under mineral deficiency of three sunflower genotypes, a population variety (Oro 9) and two hybrids (Mirasol and Albena). Sunflower seedlings were submitted to five treatments: N deficiency (N_0), P deficiency (P_0), K deficiency (K_0), N and K deficiency (N_0K_0) and a control. Plants were harvested when they reached 3-4 true pairs of leaves. Growth parameters measured (height, total leaf area, root length, root and shoot dry mater) were all significantly reduced by mineral deficiency. Leaf area was most reduced by N_0 (-61%) and P_0 (-56%). Total dry matter was most affected by N_0 (-63%) and by N_0K_0 (-66%). Genotype comparisons showed that Oro 9 had the highest shoot dry matter while Albena had the lowest root dry matter. Effect of mineral deficiency on content and partitioning of N, P, K, Ca and Na was significant and varied according to treatments and among plant parts. Shoot dry weight was significantly correlated with root N content (r^2=0.81) and root K content (r^2=-0.61) for N_0 and K_0.

Key words: **sunflower, mineral deficiency, nutrient partitioning, morphological parameters**

INTRODUCTION

Response of a plant to environmental stress often result in yield decline. Mineral deficiency induces a stress that affects plant growth, development and grain yield. Subsequent yield decrease frequently derives from disturbance of metabolic

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process at the cellular level (Kmock *et al.*, 1975). Magnitude of decline depends mainly on plant growth stage at which it occurs and on the number of deficient minerals (Salisbury and Ross, 1985; El Midaoui *et al.*, 1999). Growth stages characterized by an intense growth are the ones where needs for certain minerals are high, especially those involved in cellular division. Therefore, any deficiency at those stages will disturb organ production (Loue, 1993).

Many investigations have been carried out to study relationship between mineral deficiency and genotype biomass. Cur *et al.* (1988) and Rufty *et al.* (1984), found considerable shoot and root biomass reduction for soybean. Heitholt *et al.* (1989) and Prameswara *et al.* (1984) reported that nitrogen deficiency reduced significantly wheat dry matter but had pronounced effect on root biomass. El Midaoui *et al.* (1999) found the same response of root biomass when sunflower seedlings were submitted to N or K deficiency. Relationships between biomass and nutrient content of different plant parts were also reported. Mathers and Stewart (1982) showed that consumption of N, P, K, Ca and Mg by a plant was closely related to biomass production.

The purpose of the present work was:

1. To evaluate the effect of mineral deficiency on shoot and root growth of seedling of three sunflower genotypes induced at an early stage;
2. To describe,modifications of some physiological and biochemical parameters subsequent to mineral stress;
3. To analyze the effect of mineral deficiency on N, P, K, Ca, and Na content of stem, leaves and root growth.

MATERIALS AND METHODS

Three sunflower genotypes were tested: a variety population (Oro 9) and two hybrids (Mirasol and Albena). Characteristics of the genotypes are summarized in Table 1. Seeds of the genotypes were germinated in Petri dishes. Single seedlings were then transplanted in polyethylene pots (5.5 cm x 4.5 cm x 8.5 cm) filled with 4-mm diameter quartz sand previously washed and dried. Pots were holed at the bottom to facilitate draining.

Mineral stress treatments were obtained by applying five different nutrient solutions: lacking nitrogen (N_0), lacking phosphorus (P_0), lacking potassium (K_0), or lacking nitrogen and potassium (N_0K_0) and a complete Hoagland solution (control).

Seedlings were daily supplied with nutrient solutions. Experimental design consisted on a complete block design with three replicates.

Morphological observations

Seedlings were harvested at three to four pairs of leaves and the following parameters were measured:

Plant height (PH) was estimated as the distance from the collar to the tip of the youngest leaves;

Root length (RL) was measured from the collar to the tip of the longest root;

Shoot (SB) and root (RB) biomass were determined after separating the two parts and over drying them at 80°C until constant weight;

Total leaf area (LA) was monitored by an electronic planimeter (LiCor-3000);

Susceptibility index (IS) Susceptibility index was calculated as follow according to formula described by Maurer (1978):

IS (Shoot) = (SBC-SBT)/SBC*100

IS (Root) = (RBC-RBT)/RBC*100

where:

- SBC = shoot biomass for control, RBC = root biomass for control
- SBT = shoot biomass for treatment, RBC = root biomass for treatment**.**

BIOCHEMICAL AND PHYSIOLOGICAL PARAMETERS

Relative water content

Relative water content (RWC) was determined in adult leaves detached from three plants per genotype. Fresh weight (FW) was determined and leaves were then kept at saturated atmosphere. After 18 h incubation, leaves were gently blotted between two sheets of absorbing paper. Fresh weight of water saturated leaves (FWS) was then determined. Dry weight (DW) was determined by oven drying leaves at 80°C until constant weight. RWC was calculated by the following formula:

RWC=[(FW-DW)/(FW-FWS)]*100

Mineral nutrient determination

Nitrogen, phosphorus, potassium, calcium and sodium content of plant parts were determined as described by Benbella (1989).

We added 0.2 g of ground material in a test tube containing 2 ml of H_2SO_4 and 2 ml of H_2O_2. Tubes were heated at 200°C for 45 min. They were then removed and cool for 10 min before adding 2 ml of H_2O_2 again and reheating at 200°C for 45 min. These steps were repeated until the solution became limpid. Tubes were then removed from the heater, let to cool at room temperature and diluted 50 times with distilled water.

Nitrogen

An aliquot fraction of 0.5 ml of the diluted digestate was poured in a test tube and diluted with 4.5 ml of distilled water. A sub-sample of 0.5 ml was piped in a test tube and mixed with 5.5 ml of distilled water, 2 ml of a solution containing 0.3% nitropruside and 8.5% sodium salicylate and 2 ml of a solution containing 0.4% NaOH and 0.5% sodium dichloroisocyanurate. Tubes were gently shacken and left at room temperature for colour development. Absorbance was red at 660 nm. Nitrogen content was determined by a standard curve using solutions with known N concentration.

Phosphorus

An aliquot fraction of 1ml of the diluted digestate was mixed in a test tube with 5 ml of a vanadomolybdate solution. Tubes were shacken and kept 30 min at room temperature for colour development. Absorbance was red at 390 nm and phosphorus content was determined by a standard curve using solutions with known P concentration.

Potassium, calcium and sodium

These three cations were directly determined in the diluted digestate using a flame spectrophotometer. Contents of these elements were derived from standard curves specific to each element.

RESULTS

EFFECT OF MINERAL NUTRITION ON PHYSIOLOGICAL AND MORPHOLOGICAL PARAMETERS

Relative water content

Relative water content (RWC) of tested genotypes was significantly affected by the treatments. Genotype differences were also observed for N_0 and P_0 (Figure 1). Oro 9 had a slightly higher RWC (84.84%) than Mirasol and Albena (81.81 and 80.78%, respectively). We noticed that under N_0K_0, RWC of the genotypes remained higher than under the other treatments.

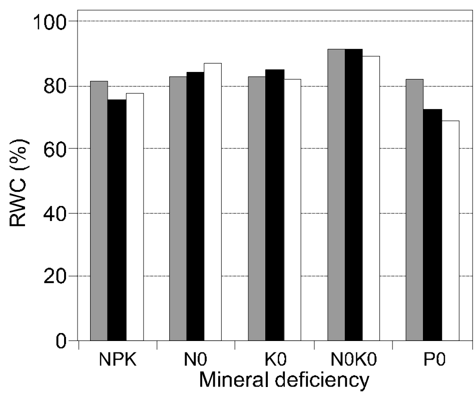

Figure 1: Effect of mineral deficiency on relative water content of three sunflower genotypes

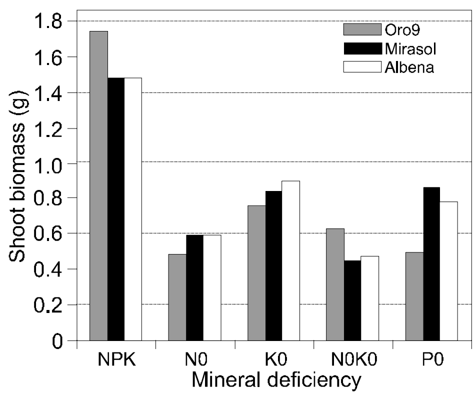

Figure 2: Effect of mineral deficiency on shoot biomass of three sunflower genotypes

Biomass

Mineral deficiency significantly reduced shoot (SB) and root (RB) biomass. Percent reduction was more than 45% for all treatments (Figure 2). Percent decline of shoot biomass varied between 46% (K_0) and 66% (N_0K_0). Declines of the same order were recorded for root biomass (Figure 3). Genotype effect on root biomass was significant for two cases; N_0 and P_0. Oro 9 appeared less susceptible to N deficiency than the two hybrids. Albena was the least susceptible to P deficiency. In the case of shoot biomass, effects were significant only for N_0K_0 and P_0. Under normal supply of nutrients, Oro 9 had a higher shoot biomass than the hybrids. Under mineral deficiency, however, significance of genotype*treatment interaction suggested that Oro 9 is most susceptible genotype to N deficiency and the hybrids most susceptible to N and K deficiencies. Effect of mineral deficiency on total biomass varied considerably among the treatments (Figure 4). P_0 and K_0 had more pronounced effects on total biomass than N_0 or N_0K_0. Oro 9 and Albena had the highest total biomass.

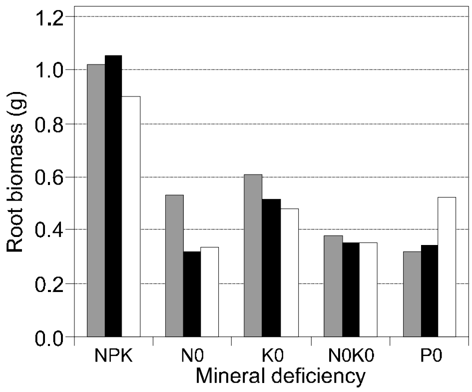

Figure 3: Effect of mineral deficiency on root biomass of three sunflower genotypes

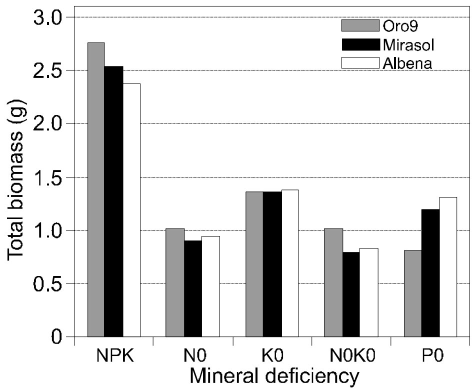

Figure 4: Effect of mineral deficiency on total biomass of three sunflower genotypes

Leaf area

Leaf area was significantly affected by mineral deficiency, genotype and genotype*treatment interaction. Leaf area showed an important decline for all treatments (Figure 5). Effects were more pronounced for N_0 (-60%) than for the other mineral deficiencies. Potassium deficiency induced the lowest decrease in leaf area (-27%). Under normal supply of nutrient, Mirasol had the highest leaf area but it declined under K deficiency.

Plant height

Under normal supply of nutrients, the genotypes did not show significant differences in plant height. Mineral deficiency significantly reduced plant height (Figure 6). Reduction was pronounced for P_0 (-22%). Nitrogen deficiency had the least effect. Differences among the genotypes were not significant. However, genotype*nutrient deficiency interaction was highly significant. Oro 9 was most susceptible to N or P deficiency.

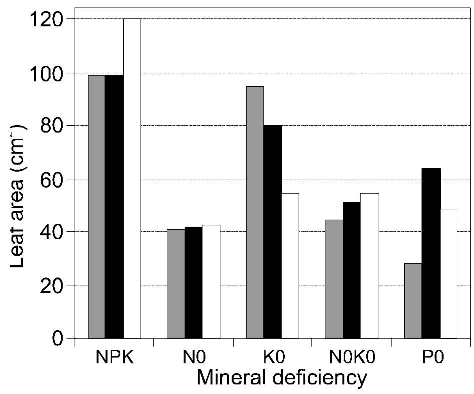

Figure 5: Effect of mineral deficiency on leaf area of three sunflower genotypes

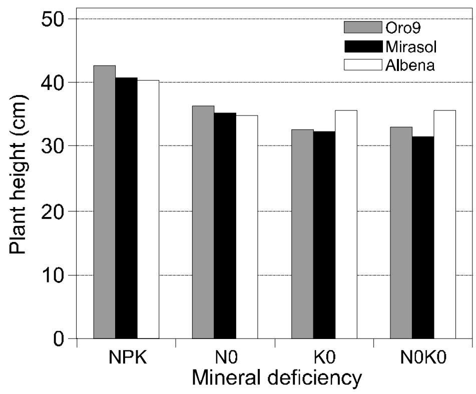

Figure 6: Effect of mineral deficiency on plant height of three sunflower genotypes

Root length

Root length was significantly reduced by mineral deficiency (Figure 7). Mirasol was the least affected.

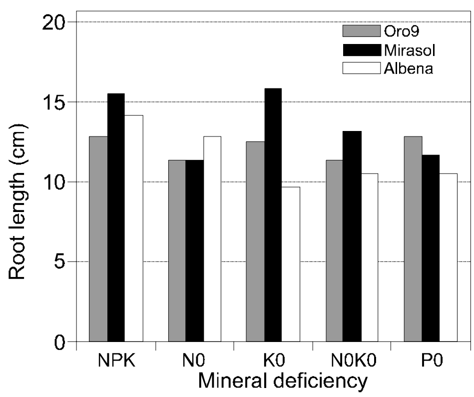

Figure 7: Effect of mineral deficiency on root length of three sunflower genotypes

EFFECT OF MINERAL DEFICIENCY ON ROOT BIOMASS/SHOOT BIOMASS RATIO AND ON SUSCEPTIBILITY INDEX

The two parameters were significantly affected by mineral deficiency (Figures 8, 9, 10, 11). Genotype differences were significant only in the case of N and P deficiencies. Average over genotypes showed that shoot biomass was more reduced under N_0 and N_0K_0 than under the other mineral deficiencies. Root biomass, however, was more susceptible for N_0. Susceptibility index showed that Albena and Mirasol the most susceptible for root biomass production and least susceptible for shoot biomass production. The ratio root biomass/shoot biomass allowed to separate genotypes in two groups; Oro 9 characterized by a high ratio as the first group and the hybrids characterized by a low ratio as the second group.

Under phosphorus deficiency, shoot biomass susceptibility index was significantly higher for Oro 9 (71.73%) than for Mirasol (45.04%) and Albena (41.26%). Root biomass susceptibility index, however, was significantly higher for Oro 9 (68.86%) and Mirasol (67.30%) than for Albena (41.26%). The ratio root biomass/ shoot biomass was higher for Albena (68.67%) and Oro 9 (62.67%) than for Mirasol (40.00%).

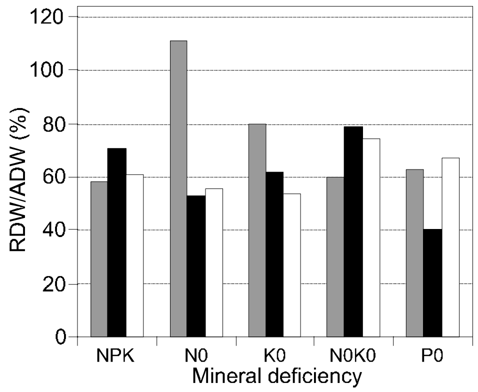

Figure 8: Effect of mineral deficiency on the ratio shoot biomass/root biomass of three sunflower genotypes

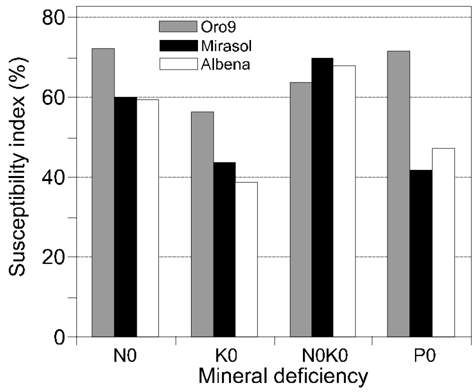

Figure 9: Susceptibility index for mineral stress calculated for shoot biomass

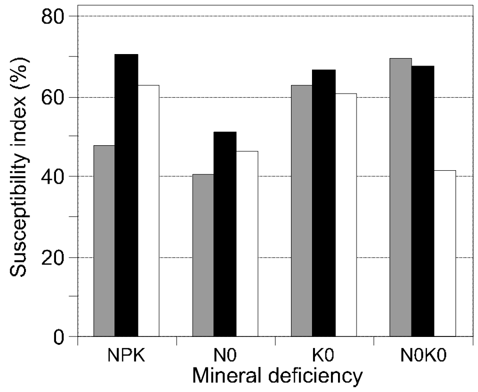

Figure 10: Susceptibility index for mineral stress calculated for root biomass

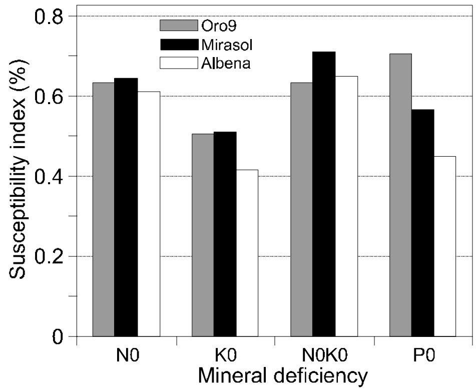

Figure 11: Susceptibility index for mineral stress calculated for total biomass

EFFECT OF MINERAL DEFICIENCY ON N, P, K, Ca AND Na CONTENT IN DIFFERENT PLANT PARTS

Nitrogen

Under normal supply of nutrients, N content, averaged over genotypes, was higher in leaves (6.1%) and roots (4%) than in shoots (1.8%). Effects of nitrogen deficiency on N content were more pronounced for stem (-45.18%) and leaves (-42.38%) than for roots (-8.95%). Combined N and K deficiencies had the same effect on the N contents of the three plant parts (Figure 12).

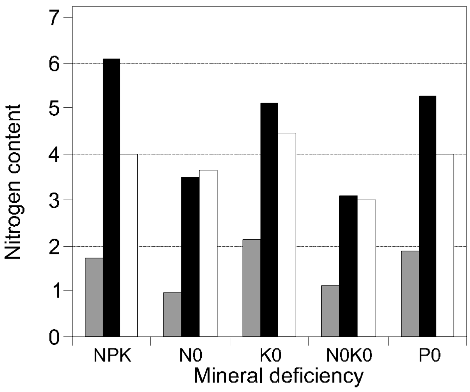

Figure 12: Effect of mineral deficiency on N content of the three plant parts of three sunflower genotypes

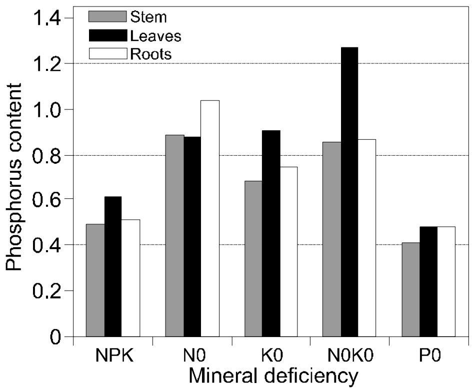

Figure 13: Effect of mineral deficiency on P content of the three plant parts of three sunflower genotypes

Phosphorus

Under normal supply of nutrients, P contents of stem, leaves and roots, averaged over genotypes, were not significant. Nitrogen deficiency significantly increased P content in roots (103%) and in stem (81%) while combined N and K deficiencies significantly increased P content of leaves (107%). Genotype differences were significant for root P content in the case of N, or K deficiencies and for N and K deficiencies. Mirasol was most susceptible (Figure 13).

Potassium

Under normal supply of nutrients, K content was high in stems (5.65%) and leaves (4.68%) and low in roots (2.8%). Among the tested genotypes, hybrids had high root and shoot K content whereas no significant differences among the genotype were found for leaves. Under mineral deficiency, the most important P content decrease was recorded for K deficiency and for combined N and K deficiencies. Stem was affected the most, followed by leaves and roots (Figure 14). Conversely, N deficiency and P deficiency increased K content in the three plant parts, but effects were pronounced for leaves (162% and 135% for N_0 and P_0, respectively) and for roots (160% and 305% for N_0 and P_0, respectively).

Sodium

Under normal supply of nutrients, average sodium content was higher in roots than in stems or leaves. All mineral deficiency treatments except N deficiency increased Na content in the three plant parts. In contrast, N deficiency reduced it (Figure 15). Sodium accumulation was pronounced in stems and roots under P deficiency, and in leaves under N and P deficiency. Genotype effect on Na content was highly significant for leaves and roots and non-significant for the stem. The hybrids accumulated more Na than Oro 9.

Calcium

Under normal supply of nutrients, leaves accumulated more Ca than the two other plant fractions. The same trend was observed under mineral deficiency (Figure 16). All nutrient deficiency treatments reduced Ca content in the plant parts, except in the case of K deficiency where Ca content increased both in leaves and stem. Comparison of treatments showed that K deficiency might induce Ca accumulation in leaves and stem and that of P accumulation might induce its accumulation in roots.

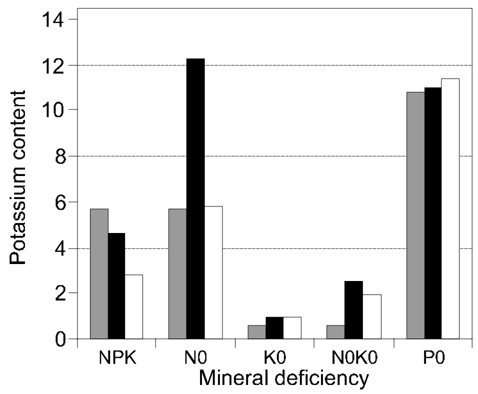

Figure 14: Effect of mineral deficiency on K content of the three plant parts of three sunflower genotypes

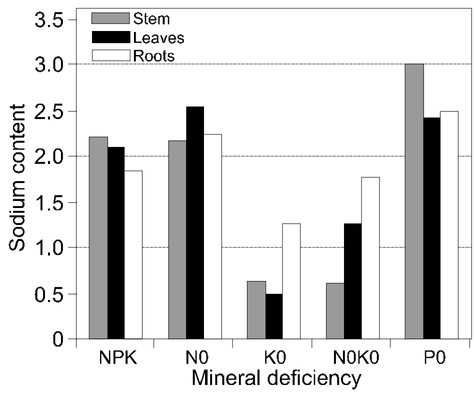

Figure 15: Effect of mineral deficiency on Na content in the three plant parts of three sunflower genotypes

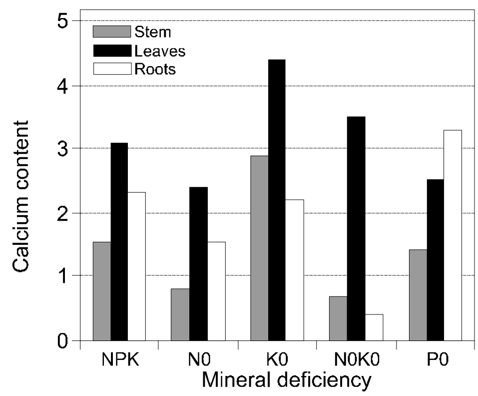

Figure 16: Effect of mineral deficiency on Ca content in the three plant parts of three sunflower genotypes

DISCUSSION AND CONCLUSION

Nutrient absorption by sunflower might be related to the level of nutrients in nutrient solution and also to biomass accumulation during early stages of development. Under normal supply with nutrients (control), seedlings of the tested genotypes behaved in the same manner, which resulted in non-significant differences for all measured parameters. However, we noticed that genotypes produced higher shoot than root biomass (1.6 times higher for shoot). In contrast, mineral deficiency reduced significantly all parameters. Effect was pronounced for plant height and for leaf area. Genotype effect as well as genotype*mineral deficiency interaction were significant for these two parameters. Percent decrease in leaf area was higher for N_0 (-61%), P_0 (-56%), N_0K_0 (-52%) but medium for K_0 (–28%). Hocking *et al.* (1987) reported identical results and found also that shoot and root biomass was significantly affected. In our case, effects on biomass were more pronounced for P_0 and K_0. Genotype differences were also significant and shoot biomass of Oro 9 was the least affected and root biomass of Albena the most reduced. El Midaoui *et al.* (1999) found similar results but reported that roots were more susceptible to N and K deficiency than shoots.

Under N deficiency, shoot and root biomass were significantly reduced but shoots were slightly more affected than roots. This result disagreed with those of Heitholt (1989) and Prameswaran *et al.* (1984) who concluded, for cereals, that shoots were less susceptible than roots. These discrepancies might be explained by growth stage at which samples were collected. Looking at biomass partitioning, considered by many workers as a mechanism of plant adaptation to environmental stress, Oro 9 in spite of a 50% shoot biomass reduction appeared to be the least susceptible to N deficiency. Shoot biomass reduction in this case, might be due to a severe decline of LA (-61%). Cure *et al.* (1988), Reed *et al.* (1988) and Rufty *et al.* (1984) reported similar suggestions.

Under normal supply of nutrients (control), sunflower seedlings had high nitrogen contents in leaves (6.09%) and roots (3.99%) and small content in stems (1.73%). This difference in nitrogen content among plant parts was also reported by Hocking and Steer (1982).

Under nitrogen deficiency, N content of plant parts declined by 45% in stems and 42% in leaves. Root showed a small decline (-9%). Shoot biomass was correlated to N content of roots ($r=0.81^{**}$) suggesting that genotypes that tolerate N deficiency are also the ones accumulating nitrogen in roots. Significant correlations between shoot biomass and K content in stems ($r=-0.79^{**}$), shoot biomass and Ca content in roots ($r=-0.70^{**}$), shoot biomass and P content in leaves ($r=-0.61^{**}$) were also observed.

Under normal supply of potassium, biomass of genotypes was not significantly different. However, under K deficiency root and shoot biomass declined drastically and percent decline was respectively 46% and 47%. Many researchers reported similar effects for different species Influence of K on osmoregulation and plant adaptation to environmental stress was also reported by several investigators (Beringer, 1978; Hocking and Steer, 1983). Nutrient partitioning, however, had received little attention from investigators. Our results showed that under normal supply of K high concentration of this nutrient was recorded in stems (5.65%), leaves (4.68%) while roots had low concentration (2.22%). Under K deficiency, plant parts showed a severe decrease in K content; 89.56% in stems, 79.28% in leaves and 66% in roots. Potassium content of roots (0.95%) remained higher than in leaves (0.79%) or shoots (0.59%). A significant correlation between K content in roots and related biomass was obtained under K deficiency (r=-0.62*). It appears that genotypes producing high root biomass are also accumulating K. Although shoot biomass did not show any significant relation with K content, it was significantly related to root biomass (r=-0.67**). Under P deficiency, content of K increased by 92%, 135% and 308% in stem leaves and roots, respectively. The same response was obtained for N deficiency only for leaves and roots. Increases of K content were 162% in leaves and 105% in roots. Significant correlations between shoot biomass and Ca content in leaves (r=0.66) and in roots (r=0.70) were obtained under P deficiency. Root biomass was also related to Ca in leaves (r=-0.81).

Under normal supply of nutrient, Na content was slightly higher in leaves than in stems or roots. In the case of Ca, leaves had the highest Ca content (2.90%) followed by roots (1.39%) and shoots (1.22%). Similar results were reported by Coïc *et al.* (1963) on vegetables and by Halbrook *et al.* (1980) on tomato. Results showed also that under K deficiency, increase in Ca content compensates for the decline in K content observed in the different plant parts. Results of Coïc *et al.* (1962) on potato and barley support this suggestion.

Sodium content decreases under K deficiency. Coïc *et al.* (1962) found the same result on potato and barley. Deficiency in both N and K reduced content of Ca and Na. Under combined N and K deficiency, Ca and Na content decreased in all plant parts. Under P deficiency, Na content increased in the three plant parts while Ca content increased in roots, remained almost constant in the stem and showed a slight decrease in leaves.

This experiment allowed to study the response of some sunflower genotypes to mineral stress by analyzing the morphological and physiological parameters as well as the susceptibility index and N, P, K, Ca and Na content. We concluded that the tested genotypes were more susceptible to N or N and P deficiency than to P or K deficiency. Susceptibility index based on shoot biomass showed that Oro 9 had higher IS under N or P deficiency than the hybrids.

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EFECTO DE LA FALTA DE ELEMENTOS MINERALAS SOBRE LOS PARAMENTROS MORFOLOGICOS Y FISIOLOGICOS Y REPARTICION DE ELEMENTOS MINERALES DE LA PARTE SOBRE EL SUELO Y DE LA RAIZ EN TRES VARIEDADES

RESUMEN

El experimento fue efectuado para investigar el comportamiento de tres genotipos del girasol, de una variedad (Oro 9) y dos hibridos (Mirasol y Albena), en las condiciones de la falta de elementos minerales. Las plantulas del girasol eran investigadas en cinco tratamientos: falta de N (N_0), falta de P (P_0), falta de K (K_0), falta N y K (N_0K_0) y control. Las plantas fueron en la fase con 3-4 pares de hojas propias. Todos los parametros investigados (alteza, superficie de hoja total, largo de la raiz, masa seca de la raiz y de la parte sobre el suelo) fueron reducidos considerablemente bajo la influencia de la falta de elementos minerales. La superficie de hoja fue reducida por lo mas con los tratamientos de N_0 (-61%) y P_0 (-56%). La masa seca total fue reducida por lo mas con los tratamientos de N_0 (-63%) y N_0K_0 (-66%). La comparacion de genotipos indico que la variedad Oro 9 tenia el valor maximo de la masa seca sobre el suelo, mientras Albena tenia el valor minimo de la masa seca de raiz. El efecto de la falta de elementos minerales sobre el contenido y la reparticion de N, P, K, Ca y Na era importante, y se cambiaba segun los tratamientos y las partes de plantas. La masa seca sobre el suelo era en la correlacion importante con el contenido de N en la raiz (r^2=0.81) en el tratamiento de N_0 con el cortenido de K en la raiz (r^2=-0.61) en el tratamiento de K_0.

INFLUENCE DE LA CARENCE D'ÉLÉMENTS MINÉRAUX SUR LES PARAMÈTRES MORPHOLOGIQUES ET PHYSIOLOGIQUES ET SUR LA DISTRIBUTION DES ÉLÉMENTS MINÉRAUX ENTRE LA PARTIE AÉRIENNE ET LA RACINE POUR TROIS SORTES DE TOURNESOL

RÉSUMÉ

Le but de l'expérience était d'examiner le comportement de trois génotypes de tournesol, une sorte (Oro 9) et deux hybrides (Mirasol et Albena), dans des conditions de carence d'éléments minéraux. Les germes de tournesol ont été examinés sous cinq traitements: carence de N (N_0), carence de P (P_0), carence de K (K_0), carence de N et de K (N_0K_0) et contrôle. Les plantes ont été récoltées à la phase de 3-4 feuilles véritables. Tous les paramètres examinés (hauteur, surface feuillue totale, longueur de la racine, masse sèche de la racine et de la partie aérienne) étaient diminués de façon importante dans les conditions de carence d'éléments minéraux. La surface feuillue a été le plus significativement diminuée par les traitements N_0 (-61%) et P_0 (-56%). La masse sèche totale a été le plus significativement diminuée par les traitements N_0 (63%) et N_0K_0 (-66%). La comparaison des génotypes a montré que la sorte Oro 9 avait la plus grande valeur de masse sèche de la partie aérienne alors que l'Albena avait la valeur la plus faible de la masse sèche de la racine. L'effet de la carence en éléments minéraux sur le contenu et la distribution N, P, K, Ca et Na s'est montrée importante et changeait selon les traitements et les parties de la plante. La masse sèche de la partie aérienne était en corrélation importante avec le contenu de N dans la racine (r^2=0.81) dans le traitement N_0 et avec le contenu de K dans la racine (r^2=-0.61) dans le traitement K_0.