Article

Nutrient deficiencies induced in fig tree cv. Neza in hydroponic conditions

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Abstract

The fig tree (*Ficus carica* L.) is one of the oldest trees in the world, produces fruits with high nutritional content and is used for medicinal purposes. Currently, this crop has been especially important in Mexico, that is why this research was aimed at: analyzing the nutritional deficiencies induced in fig plants *Ficus carica* cv. Neza in hydroponic conditions using the missing element technique. To carry it out, six solutions were prepared, one with all the ions and five with a missing nutrient element. The experiment was carried out under a completely randomized design with six treatments (complete solution and five of them without a macronutrient: nitrogen, phosphorus, potassium, calcium and magnesium) with five replications. Growth variables were evaluated: number of leaves, height, stem diameter, number of fruits and leaf area. In addition, chemical tissue analyzes were performed to confirm nutritional deficiency and visual symptoms. Data were analyzed through ANVA and Tukey tests ($\alpha = 0.05$). Significant differences were detected between the treatments without nitrogen and without calcium with respect to the complete solution (Solcomp) in the variables of growth and nutritional content, manifesting visually. In the treatment without phosphorus (SP) there were no significant differences. In conclusion, the information on the content of N, P, K, Ca and Mg in the leaf tissue will serve as a reference to producers of this species to correct nutritional deficiencies of these elements in the development of the fig tree.

Keywords: *Ficus carica* cv. Neza, foliar analysis, physiopathy, visual symptoms.

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Introduction

The research presented is the result of a series of experiments proposed and elaborated using the missing element technique in hydroponic conditions according to the methodology of Fernández-Pavia *et al*. (2015) started with the cultivation of prickly pear and continuing with different fruit trees and strawberries. The current work shows the results in the fig plant *Ficus carica*, cv. Neza. The fig tree (*Ficus carica* L.) is one of the oldest fruit trees. Simão (1998) mentioned that its origin is located in Asia Minor (Turkey) and Lev-Yadun *et al*., 2007) mentioned that it was domesticated approximately 6 500 years ago.

The fig tree is a plant that stands out for producing fruits with a high nutritional content, with sugars, minerals, antioxidants and vitamins A, B and C, with a high content of calcium, iron and copper. The ripe fruit is consumed dry, raw, pickled or in jam, it is used for medicinal purposes as an anticancer, laxative, anthelmintic, painkiller, cough remedy or certain skin infections (Hashemi and Abediankenari, 2013). The fig tree belongs to the *Moraceae* family and more than 750 varieties are known (López-Corrales *et al*., 2011) in all the warm regions of the world. FAOSTAT (2018) published that the four main fig-producing countries are Turkey (306 499t), Egypt (189 339t), Morocco (128 380t) and Algeria (109 214t).

Muñoz-Villalobos *et al*. (2015), explained that, in Mexico, fig trees have persisted since colonial times, in the form of creole populations in diverse ecological regions, with little agronomic management. Mendoza-Castillo *et al*. (2017) considered that currently the cultivation of this species has increased as an economic alternative for the rural sector. They also explained that the United States of America imports more than $11 million annually of fresh and processed figs, from Turkey and Italy.

SAGARPA (2018) published that, in Mexico, there is an area planted with fig trees of approximately 1 357.75 ha, with an annual production of 7 704.98 t. The entities that participate in fig production in order of importance are: Morelos (39%), Baja California Sur (22%), Veracruz (16%), Puebla (12%), Hidalgo (4%), Durango (2 %), Jalisco (2%), Mexico City (1%), San Luis Potosí (0.4%), Sonora (0.4%) and Zacatecas (0.1%).

Rivera *et al*. (2017) explained that the information that exists worldwide, is focused on fertility aspects, and highlights that the levels of nitrogen and potassium promote the highest harvest rates. In Mexico, so far, there is little information in documentary sources about visual symptoms induced by nutritional deficiencies in the fig tree. Therefore, the following research question was asked: How are nutritional deficiencies manifested in fig plants *Ficus carica* cv. Neza induced under hydroponic conditions using the missing element technique?

To answer this question, an investigation was proposed that aimed to analyze the nutritional deficiencies induced in fig plants *Ficus carica* cv. Neza under hydroponic conditions using the missing element technique. The hypothesis proposed was that: Using the missing element technique under hydroponic conditions, it is induced in the fig plant *Ficus carica* cv. Neza nutritional deficiencies that are visually noticeable and detected in chemical analysis.

Materials and methods

Experiment location

The investigation was carried out under greenhouse conditions, in the Dr. Ramón Fernández González Research Unit located within the coordinates 19º 46’ north latitude and 98º 88’ west longitude and at an altitude of 2 250 m, located in Lomas de San Esteban, Texcoco, State of Mexico.

Vegetal material

A creole fig cultivar *Ficus carica* cv. Neza, from backyard orchards propagated in the Postgraduate College, *Campus* Montecillo. The cultivar is early since it was harvested five months after the rooting of the stake. The fig tree has pale black epicarp fruits and purple flesh.

It is characterized by being vigorous and having a great environmental adaptation at altitudes between 1 000 and 3 000 meters above sea level and the fruit develops optimally in temperatures ranging between 12 and 40 °C. The fruit is parthenocarpic with an average size (72.9 ±20.2 g), black color at its maximum maturity, those without total soluble solids average 17.3 ±5.4 °Brix, its shape is slightly elongated with a length/width ratio of 1.41 ± 0.3 and it can be stored at 4 ±1 °C for 15 days without detracting from its quality.

Obtaining fig plants

Stakes cut from a seven-year-old tree four meters high were used. They were cut of 25 cm in length, treated in the basal part with indolebutyric acid potassium at a concentration of 200 ppm. Rooting was carried out for 90 days in a 500g black plastic container using peat as a substrate.

Nutritive solution

The composition of the nutritional solutions used in the experiment was based on Steiner’s (1966) universal solution for macronutrients (me L^{-1}. NO_3^- 12, $H_2PO_4^-$ 1, SO_4^{2-} 7, K^+ 7, Ca^{2+} 9, Mg^{2+} 4). The concentration of micronutrients (mg L^{-1}) for all the solutions was: Fe 4, B 0.87, Mn 1.6, Zn 0.23 and Cu 0.011 according to that proposed by Fernández-Pavia *et al*. (2012).

Establishment of the experiment

The experiment was established between the months of April to July 2017. The root stake was removed from the root ball substrate and transplanted into a closed hydroponic system, which consisted of individual 6 L plastic containers, painted in black color with perforated cover; each container was fitted with a 4 mm diameter hose with forced air, to oxygenate the roots with levels of at least 2 m gL^{-1}. Each container was considered as an experimental unit, which was adjusted to a volume of 5 L with universal nutrient solution (Steiner, 1966 in Fernández-Pavia *et al*. 2012). The solution was prepared with reactive grade chemical salts, the pH of the solution was adjusted between 5.5 and 6, with a Conductronic® brand pH meter.

Experimental design and treatments

After the adaptation period, the application of the ‘missing element technique’ began. For this, six treatments were established and six solutions were prepared, one with all ions (complete solution-SCOMP and five with a missing nutrient element in fig plants to induce symptoms related to deficiencies corresponding to N, P, K, Ca, and Mg, with five repetitions per treatment, which were distributed under a completely randomized experimental design.

The plants underwent treatments for 90 days between the months of May and July 2017. During the 90 days, the volume of solution spent in each experimental unit was restored with distilled water to the original volume of 5 liters and adjusted pH. Likewise, the nutritional solutions were completely renewed every 15 days.

Variables evaluated

The variables evaluated were number of leaves, number of fruits, plant height (cm), stem diameter (cm) and LI-COR© leaf area (cm^2). Also, the content of N, P, K, Ca and Mg in leaves (g kg^{-1}) was determined.

N, P, K, Ca and Mg content (g kg^{-1})

It was determined by means of chemical analysis of tissue in 90-day-old plants. All the leaves of each plant were sampled, placed in paper bags and dried in a Thermolab© forced air oven for 24 h at 70 °C. Once dry, the samples were ground in a ©Thomas scientific 5 mm mesh stainless steel sieve for a Wiley© stainless steel mill, fitted with a 40 mesh sieve. With this material, the total nitrogen content was quantified, using the semi-micro-Kjedahl method, described in the AOAC manual (1995). The determination of P, K, Ca and Mg were carried out by means of wet acid digestion (AOAC, 1995) and quantified by ICP- AES Varian®.

Symptomatology

The recording of the symptoms of deficiencies in leaves was carried out visually and expressed by photographs (Figures 1, 2, 3, 4, 5). Plants were observed every third day to record the incidence of chlorosis, presence of yellow, reddish or purple spots, necrosis, deformities, decreased growth or death of plants.

Analysis of data

To begin, the data obtained from values reported by two different authors who worked on the macronutrient contents of the fig tree were contrasted with those obtained in this work. Then, the data obtained through Shapiro-Wilk normality tests (α= 0.05) were analyzed, where it was observed that all the variables behaved under the normal distribution. Levene tests (α= 0.05) were also applied to verify homoscedasticity. Later, analysis of variance (ANVA) and Tukey means comparison tests (α= 0.05) were carried out for both growth parameters and nutritional content of the leaf tissue. The calculations were made supported by the SAS 9.4 package under Windows.

Results and discussion

In Table 1, the adequate values of N, P, K, Ca and Mg for the fig culture are presented according to Raij *et al*. (1996) and Malavolta (1997) that were reported by Leonel and Costa (2011). Also, the values obtained in this work are shown when there is a macronutrient absence of any element in leaf tissue.

Table 1. Values of macronutrients considered adequate in the cultivation of fig (*Ficus carica* cv. Neza) against those obtained in this investigation with the missing element.

Macronutrient	Raij *et al*. (1996)	Malavolta *et al*. (1997)	Values obtained
N	20-25	22-24	18.97
P	1-3	1.2-1.6	1.7
K	10-30	12-17	0.761
Ca	30-50	26-34	10.05
Mg	7.5-10	6-8	1.27

The values are expressed in g kg^{-1}.

In the Table 1 highlights that only in phosphorus (P) there were no nutritional deficiencies. Later, the behavior of each of the elements is explained in greater detail. Also, the results obtained both in the ANVA and in the Tukey mean comparison test (α= 0.05) of growth variables in fig plants *Ficus carica* cv. Neza. Table 2 shows the results obtained both in the ANVA and in the Tukey mean comparison test (α= 0.05) of the macronutrient content in the leaf of the fig cv Neza.

Table 2. Comparison of means of growth variables in fig plants *Ficus carica* cv. Neza.

Treatment	No. of leaves	Plant height (cm)	Stem diameter (cm)	No. of fruits	Leaf area (cm^2)
SCOMP	15.6 ab	31.54 a	0.95494 ab	7.4 a	1218.8 a
SN	1.2 c	9.92 c	0.66526 cd	0 b	15.4 b
SP	16.8 a	26.52 abc	0.98042 a	6 a	1259.8 a
SK	14.8 ab	27.88 ab	0.91038 abc	6.8 a	570.9 b
SCa	1.4 c	13.61 bc	0.6016 d	0.2 b	48.6 b
SMg	8.2 bc	14.3 bc	0.69392 bcd	1.4 b	302.7 b
DMS	7.9793	17.227	0.2625	3.3397	595.57
CV	42.21	42.7	16.75	47	51.44

SCOMP= solution with all the nutrients; SN= nitrogen-free solution; SP= phosphorous-free solution; SK= potassium-free solution; SCa= calcium free solution and SMg= magnesium-free solution; DMS= minimal significant difference; CV= coefficient of variation in percentage. Averages with different letters in the same column are statistically different, according to the Tukey test ($p \leq 0.05$).

In Table 2, lower values were distinguished in number of leaves, plant height, stem diameter, number of fruits and leaf area in the solutions without nitrogen, without calcium and without magnesium. Not extremely high coefficients of variation were detected in most of the variables. Garza-Alonso *et al*. (2019) detected the same behavior in the variables plant height and stem diameter for the cases of solutions without nitrogen and without calcium in another fig cultivar. Table 3 shows the averages of macronutrient concentrations obtained from the fig leaves *Ficus carica* cv. Neza and Tukey's mean comparison test.

Table 3. Macronutrient concentrations in fig plant leaves *Ficus carica* cv. Neza.

Treatment	N	P	K	Ca	Mg
SCOMP	25.743 ab	2.01 b	7.439 bc	26.732 ab	4.7064 bc
SN	18.969 b	2.6244 ab	13.174 a	13.362 cd	2.8142 cd
SP	24.213 ab	1.7066 b	6.131 c	22.562 b	3.865 c
SK	31.44 a	3.6756 a	0.761 d	32.657 a	8.4498 a
SCa	23.642 ab	2.0668 b	10.432 ab	10.052 d	6.4608 ab
SMg	27.642 ab	2.9708 ab	13.007 a	19.087 bc	1.2752 d
DMS	11.095	1.488	3.934	8.95	2.536
CV	22.44	30.33	23.69	22.06	28.21

The data in the table is in g kg^{-1}; SCOMP= solution with all the nutrients; SN= nitrogen-free solution; SP= phosphorous-free solution; SK= potassium-free solution; SCa= calcium free solution and SMg= magnesium-free solution; DMS= minimal significant difference; CV= coefficient of variation in percentage. Averages with different letters in the same column are statistically different, according to the Tukey test ($p \leq 0.05$).

In Table 3 it was detected that the solution without nitrogen has an antagonistic effect with potassium. When eliminating phosphorus (SP), the behavior of the different macronutrients was similar to that of the complete solution (SCOMP). By eliminating potassium (SK), the absorption of phosphorus was increased due to an antagonistic effect of these ions and the calcium concentration increased due to a synergistic effect. The calcium-potassium and magnesium-calcium effects have an antagonistic relationship. The same behaviors coincide with those reported by Nuñez-Escobar (2016).

Nitrogen

In relation to the solution without nitrogen (SN), the variables number of leaves, plant height, stem diameter, number of fruits and leaf area were affected (Figure 1a), since the plants had almost no growth, with little number of leaves, generalized chlorosis, sparse foliage, as well as a plant with a stunted and yellowish appearance (Figure 1b).

This could be verified both in the ANVA and in the Tukey means comparison tests, where highly significant differences were detected in the growth variables when compared with the complete solution (SCOMP) as shown in Table 2. In the chemical tissue analysis, it was confirmed that the N content value was the lowest, a significant difference was also detected with the treatment without potassium (SK) and not significant with respect to the rest of the treatments (Table 3).

a) b)

Figure 1. Visual symptoms of nitrogen deficiency (SN), a) initial stage; b) final stage and comparison with the complete solution.

Plants subjected to the lack of this element, after a month of starting the experiment, decreased their growth rate (Figure 1a). The leaves presented a light green color after 15 days, later accentuating it until it became chlorosis, the symptoms in the final stage were evident throughout the plant, due to the absence of this element, the plants had fewer leaves or no leaves stopped their growth (Figure 1 b).

These symptoms coincided with those observed by Rice (2007), who mentioned that, under conditions of nitrogen deficiency, the plants show slow growth, with a reduced size, thin branches in smaller numbers and with a tendency to vertical growth, fewer leaves, which reduces the leaf area, generalized chlorosis and premature leaf fall. Marschner (2012) reported that, in several crops, the initial and most severe symptoms are seen in the most senescent leaves where proteins hydrolyze and N is translocated as amino acid from old tissues to the actively growing portions of the plant.

Proteolysis results in a collapse of the chloroplasts, with a decrease in the chlorophyll content and the yellowing of the leaves (Marschner, 2012). This phenomenon could have occurred in fig trees, since the most severe symptoms were observed in more senescent leaves. Marschner (2012); Alcántar and Trejo (2012) explained that N deficiency limits cell division and expansion, as well as the development of chloroplasts, since it alters metabolic processes in crops, which produces weak, stunted, slow-growing plants that mature early and with low yield. This is in agreement with what was observed in this study, since stress due to the lack of this macronutrient significantly affected all growth variables.

Phosphorus

The relationship of the growth variables in the solution without phosphorus (SP) are shown in Table 2. In the Tukey test, no differences were detected with ($p\leq 0.05$) between the treatment without phosphorus (SP) and the complete solution (SCOMP). In relation to the content of 1.7

g kg^{-1}, the phosphorus concentration is within the range reported by Raij *et al*. (1996) of 1-3 g kg^{-1} but not within those of Malavolta (1997), who reported a content of 1.2 to 1.6 g kg^{-1}, as shown in Table 1. Regarding visual symptoms In Figure 2B, a slight reduction in growth was observed, unlike that of the complete solution (SCOMP).

Also, leaves with a green of similar shades were detected in both treatments. With the above, it can be inferred that this response was due to the fact that the reserves of the plant were sufficient in the evaluated period of 90 days. Garza-Alonso *et al*. (2019) distinguished some deficiencies in fig trees from another cultivar at 120 days. Likewise, in other crops, phosphorus deficiency is reported by the presence of anthocyanins, causing the leaves to turn purple (Fernández-Pavia *et al*., 2015).

a) b)

Figure 2. Visual symptoms of phosphorus deficiency. a) initial stage; b) final stage and comparison with the complete solution.

Potassium

In the ANVA (Table 2) it was distinguished that at least one of the means of a treatment were different. When contrasting all the values of the solution without potassium (SK) against the complete solution (SCOMP), only differences in means were observed in the Tukey test in the leaf area, the rest were statistically similar. The visual symptoms of potassium deficiency were one of the last to become evident, until day 71 after undergoing treatment without Potassium (Figure 3). It was observed that in the initial stage the leaves showed curl of the margins inwards and with necrosis in the mature leaves.

The deficiency of this element was manifested in adult leaves with necrotic areas towards the central part of the leaves, in the shape of a ‘V’. Salazar-Garcia (2002) pointed out that the curvature of the leaves is due to a response of the plant to avoid water loss and compensate for stoma malfunction. Potassium is involved in photosynthesis, since it regulates the opening and closing of stomata and therefore regulates the absorption of CO_2 and triggers the activation of enzymes and is essential for the production of adenosine triphosphate (ATP).

a) b)

Figure 3. Visual symptoms of potassium deficiency, a) initial stage; b) final stage and comparison with the complete solution.

The treatment without potassium (SK) was the one with the highest calcium content (32 657 g kg^{-1}) due to its antagonistic effect with this ion, compared to the other treatments, and it is within the levels reported for the cultivation of fig (Table 1). The observed symptoms could be related to this physiological involvement. Regarding the concentration found in the chemical analysis (Table 3), significant differences were distinguished with the rest of the solutions.

Rivera *et al*. (2017) and Leonel and Tecchio (2009) agreed that the nutrients required in greatest quantity by the fruits of the fig tree are nitrogen and potassium. In the treatment without nitrogen (SN) there are no fruits and in the one without potassium (SK) there are, in the same way as those of the complete solution (SCOMP), but the quality of the fig is unknown. Ramírez *et al*. (2009) mentioned that potassium deficiency increases calcium values, since they compete for absorption sites in the roots, therefore K deficiency can accentuate Ca abundance as well as for N and P.

Calcium

In the ANVA and in the Tukey means comparison tests (Table 2), highly significant differences were distinguished between the complete solution (SCOMP) and the calcium-free solution (SCa) in all the growth variables. Calcium deficiency is shown in Figure 4. In the initial stage chlorosis of the tips and tips of young leaves was detected, deformation of the leaves generally hooked downwards and often chlorosis in the new growth. In the final stage, it was associated with general chlorosis, premature leaf drop, and a decrease in stem diameter that was similar to that presented in the solution without nitrogen (SN).

What was obtained coincided with that reported by Marschner (2012), who mentioned that calcium deficiencies are manifested, with a lower capacity for protein synthesis in plants, which influences less radical development, marked chlorosis in leaves (mainly young) and little growth of stems and leaves. Likewise, the visual symptoms coincided with that reported by Epstein and Bloom (2016) for other crops where the whitish-yellow lesions increase in size, and in the case of very severe deficiency, the new leaves die as soon as they appear.

a) b)

Figure 4. Visual symptoms of calcium deficiency, a) initial stage; b) final stage and comparison with the complete solution.

Magnesium

The magnesium-free solution (SMg) presented lower values than those obtained with respect to the complete solution (SCOMP) for the values of height, number of fruits and leaf area. In the ANVA and the Tukey tests the differences were highly significant (Table 2). In the foliar tissue concentration, it was observed that there are highly significant statistical differences with respect to treatment without K (Table 3).

Based on the visual symptomatology (Figure 5), in the initial stage a yellowish color was detected between the ribs, the mature leaves being the most affected, in the final stage it was observed that the height of the plant is different and the behavior shown in the initial stage is very noticeable until the leaves fall.

a) b)

Figure 5. Visual symptoms of magnesium deficiency, a) initial stage; b) final stage and comparison with the complete solution.

In a study carried out by Leonel and Tecchio (2009), when evaluating the content of nutrients in leaves and fruits of the cultivar Roxo de Valinhos, they concluded that there was no limitation of nutrients for the growth and production of this fruit; however, the Mg content was below the level recommended by Raij *et al*. (1996, in Leonel and Costa, 2011).

Conclusions

The objective in this investigation was fulfilled. The proposed hypothesis is not rejected because the technique used under hydroponic conditions allowed to detect nutritional deficiencies in fig plants *Ficus carica* cv. Neza. It was highlighted that in the growth variables in the treatment without phosphorus (SP) there are no significant differences with respect to the complete solution (SCOMP), in other nutrients N, P, K, Ca and Mg if differences were detected both in statistical values as visually.

The information on the content of N, P, K, Ca and Mg in the foliar tissue will serve as a reference for producers to be able to correct nutritional deficiencies of these elements in the cultivation of fig *Ficus carica* cv. Neza. The rigor of this research -in terms of theoretical foundation, manufacturing process, chemical tissue analysis and statistical tests- make the proposal sustainable over time. This experience is transferable to other crops or other fruit trees in contexts similar to that of this experiment, inducing nutritional stress under greenhouse conditions.

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