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Symptoms of Nutrient Deficiencies on Cucumbers

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Authors' contributions

This work was carried out in collaboration between all authors. Authors VVC and LCC are postgraduate students and made the literature searches, conducted the experiment, and wrote the first draft of the manuscript. Author ABCF is adviser of the students, and designed the study, the materials and methods item, managed the experimental process, and helped them in the text editing.

All authors read and approved the final manuscript.

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ABSTRACT

Aims: The objective of this study was to describe and photograph the initial symptoms and the development of nutritional deficiencies in cucumbers, and determine the concentration of the omitted macronutrient in leaves, on which symptoms of deficiency were observed.

Study Design: A completely randomized design with six treatments and three replications were made.

Place and Duration of Study: Sector of Horticulture Crops and Aromatic Medicinal Plants belonging to the Department of Plant Production of the Faculty of Agriculture and Veterinary Sciences, UNESP Jaboticabal, São Paulo, Brazil, from March 6 to May 9, 2014.

Methodology: Treatments corresponded to a complete nutrient solution and to individual omissions of N, P, K, Ca and Mg, when plants were in early fruiting. The symptoms of macronutrient deficiencies were described and recorded by photographs, and the omitted macronutrient contents were determined at the onset of symptoms.

Results: Physiological deficiencies of N, P, K, Ca and Mg in cucumber were first observed at 3, 20,

2, 3 and 5 days after nutrient omission, respectively. N and P deficiencies were initially characterized by uniform chlorosis and browning, respectively, while the lack of K lead to chlorosis initially on the margins of the leaf blade progressing to necrosis. The Mg deficiency was characterized by internerval chlorosis, which evolved to tissue necrosis. Ca deficiency symptoms were observed in leaves next to meristems, with leaf shriveling and marginal tissue necrosis of the leaf blade. At the observation of deficiency symptoms, the levels of N, P, K, Ca and Mg in leaves with a deficiency were 18.9, 1.5, 9.5, 1.5 and 1.2 g kg⁻¹, respectively.

Conclusion: With the omission of nutrients in early fruiting plants, the symptoms of the deficiencies of N, P, K and Mg are observed initially in intermediary leaves and Ca deficiency in young leaves.

Keywords: Cucumis sativus L; nutritional lack; nutritional deficiency; plant nutrition.

1. INTRODUCTION

Production systems need to reduce costs, conserve natural resources and minimize without environmental impact sacrificing productivity and quality of crops. This can be achieved through a proper fertilization planning [1], mainly with vegetables, since this cultural practice accounts for almost 50% of cucumber (Cucumis sativus L.) production costs [2], besides being fundamental to the success of farms. Therefore, the provision of information concerning plant nutrition bases, such as identifying nutritional deficiencies, will assist farmers facing decisions on planning future fertilizations.

When the supply of nutrients is inadequate for the normal growth and development of plants, metabolic disorders occurs [3]. They give rise to deficiency symptoms [4], which cause yield decline and poor quality of cucumbers [5,6]. In addition, they are detrimental to both horticulture and the environment. Therefore, the assessment of plant's nutritional status becomes a valuable tool to try to predict plant's growth, development, productivity and quality by identifying the limiting element [7].

To evaluate the nutritional status of plants, a visual diagnosis of nutritional deficiencies stands out among the techniques used on crops because it is simple and can be used directly in the field. However, it has some limitations, such as symptoms being mistaken for other deficiencies caused by other biotic or abiotic stress agents [1]. They may also appear differently, with peculiarities according to species, level of missing nutrient or crop's growth stage [8], which may make it difficult to be diagnosed by farmers. Thus, a foliar analysis is needed to complement and confirm visual diagnoses.

In this context, identification, detailed descriptions and photographs of nutrient deficiency symptoms accompanied by leaf content analyses will provide valuable information, which may help horticulture farmers with crop's nutritional diagnoses and with facing decisions for a proper fertilization planning.

The objective of this study was to describe and photograph the initial symptoms and the evolution of macronutrient deficiencies in cucumbers, and verify the macronutrient content omitted from the leaf on which a symptom of deficiency was observed.

2. MATERIALS AND METHODS

2.1 Plant Material and Growth Conditions

The experiment was conducted in a greenhouse under hydroponic system - *nutrient film technique* (NFT) at the Sector of Horticulture Crops and Aromatic Medicinal Plants belonging to the Department of Plant Production of the Faculty of Agriculture and Veterinary Sciences, UNESP Jaboticabal, São Paulo, Brazil (21°15'22"S, 48°8'58' W, 575 m of altitude), from March 6 to May 9, 2014.

Omissions of nitrogen (- N), phosphorus (- P), potassium (- K), calcium (- Ca) and magnesium (- Mg). Each experimental unit consisted of a growing channel with four cucumber plants.

The sowing of 'Nikkey' Japanese cucumber was held in a phenolic foam with 54 cells (5 x 5 x 3 cm). It remained in a greenhouse for germination, emergence and early seedling growth until cotyledon leaf expansion. Afterwards, the seedlings were transferred to NFT hydroponic channels with 5 cm in diameter. When they had two leaves, they were transplanted to hydroponic definitive channel.

The definitive cultivation channels corresponded to PVC pipes, 0.2 m diameter and 2 m length, cut along the greater diameter of the tube and covered with Tetrapak[®] to avoid the incidence of solar radiation in the nutrient solution. The spacing between plants was 1.1 m between cultivation channels and 0.5 m among plants in channels.

Until the application of the treatments, the plants received a complete nutrient solution [9], whose nutrient concentrations, in mg L⁻¹, were 14, 196, 31, 234, 200, 46, 64, 5.2, 0.57, 0.51, 0.02, 0.11 and 0.01 of N-NH₄⁺, N-NO₃⁻, P, K, Ca, S, Mg, Fe, Mn, B, Cu, Zn and Mo, respectively. Nutrient solutions from omission treatments were formulated aiming to omit the respective macronutrient for each treatment without changing the concentration of other nutrients. To do so, ammonium nitrate, potassium nitrate, nitrate. magnesium calcium nitrate. phosphate, monobasic monoammonium potassium phosphate, potassium sulfate. magnesium sulfate, calcium chloride, potassium chloride, potassium acetate, calcium acetate, nitric acid, sulfuric acid, copper sulfate, zinc sulfate. sulfate. manganese ammonium molybdate, boric acid and Fe-EDDHMA were used.

At 38 days after sowing, when plants were in early fruiting, the treatments were applied with the replacement of the complete nutrient solution for solutions with the omission of one of the macronutrients. An exception was made to K, because it is the most accumulated nutrient in cucumbers and because it is observed in high amounts [10], whose nutrient concentration in the nutrient solution was initially reduced to 10% of the total present in the complete solution without symptoms of deficiency being observed, Therefore, the nutrient was ommitted in the nutrient solution. Because hydroponic water supply contains 19 mg L⁻¹ Ca, deionized water was used in the treatment with Ca omission.

Each plot consisted of an independent NFT system with a 150 L reservoir of nutrient solution, which were pumped by a submerse pump (Chosen, Power Head CX-300, with a flow rate of 1,000 L h⁻¹) to the head of the channel by a ³/₄ inch hose. The pumps were controlled by a timer allowing the circulation of the nutrient solution during the day, from 6am to 18 pm, without interruption. Cucumber plants were vertically supported by plastic strip; these plants had one main vertical stem, without sprout thinning of

lateral branches aiming the greatest leaf area, and increase in the demand for nutrients and the possibility of occurrence of nutritional deficiency symptoms were sought for analyses.

The pH of nutrient solutions was measured every two days with the use of a portable digital pH meter (Hanna, model HI 98108) and kept between 6.0-6.5 using sulfuric acid and sodium hydroxide. Due to pH 8.2 of the hydroponics water supply, when the nutrient solutions were prepared or when water was added to restore the level of the reservoir, the pH was adjusted with the use of portable digital pH meter (Hanna HI model 98108) and kept between 6.0-6.5 using sulfuric acid. The electrical conductivity (EC) was kept between 1.8 and 2.2 dS m⁻¹ using a portable digital conductivity meter (Hanna, model HI 98312), which was restored to its initial value whenever a 20% reduction of the initial EC was found. A nutrient stock solution with the same concentration of the initial solution of nutrients was used for each treatment stored in polyethylene containers with a capacity of 1,000 L. At every three EC replacements with the nutrient stock solution, approximately every 15 days, the nutrient solutions were completely renewed.

2.2 Determination of Leaf Nutrient Content

Symptoms of macronutrient deficiencies were described and recorded by photographs. To evaluate nutritional status, the recently-mature whole leaf in the early fruiting [1] was used in plants grown with the complete nutrient solution. this treatment, the levels of all the macronutrients were determined. In other treatments, omitted macronutrient contents were determined at the onset of symptoms, according to methods described by [11]. To do so, leaves were classified as old leaves (OL; situated between the third and the fifth node from the base of the plants); intermediary leaves (IL; located between the fifth and the eighth node from the tip, excluding the apical tuft); and new leaves or leaves near meristems (NL: located from the third node from the tip, excluding the apical tuft).

3. RESULTS AND DISCUSSION

3.1 Nitrogen

The onset of N deficiency symptoms came three days after the omission of this nutrient in the

nutrient solution. It was characterized by a slight loss of blade's green hue of intermediary leaves in the main stem (Fig. 1). The rapid and continuous growth of the crop until harvest [10], plus the quantity demanded and the importance of the nutrient in plant metabolism [4,7], explain the celerity of the emergence of deficiency symptoms.

At this time, the levels of new, intermediary and old leaves was 33.4, 18.9 and 20.5 g kg⁻¹ N, respectively (Table 1), characterizing a redistribution of this nutrient in the plant. According to [1], cucumber plants that have N deficiency show a foliar content of less than 25 g kg⁻¹ at earlier fruiting. Therefore, upon observing initial symptoms (at the 42nd day of the cycle), only new leaves had an adequate N content.

Both in plants cultivated with a complete nutrient solution (CS) and in plants with N omission, the

nutrient content increased in favor of the meristematic region, although differences between the contents of old and new leaves were lower (12.2%) than among leaves of deficient plants (38.6%) (Table 1). The N content in the standard nutritional status evaluation sheet of plants grown in CS was 57.8 g kg⁻¹, greater than the adequate range (25-50 g kg⁻¹) suggested for the culture [1], and lower than intermediary leaves of deficient plants (Table 1).

Chlorosis of old leaves originates after the nutrient redistribution process [12], since proteolysis of the Rubisco enzyme and other proteins from the chloroplast occurs, releasing the nutrient shaped into amino acids to meet the demand of new organs [13] aiming to ensure growth and development. Considering this, the lower level of difference between old and new leaves on plants grown in CS other than -N plants is also explained.

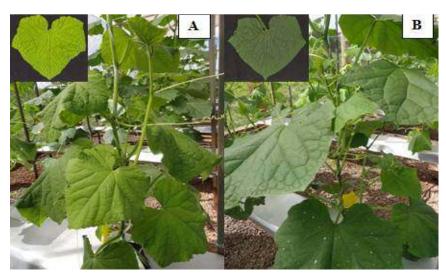


Fig. 1. Cucumber plant with chlorosis in intermediary leaves due to the omission of N (A) compared to plants grown in complete nutrient solution (B)

Table 1. Content (g kg⁻¹) of leaf nitrogen, phosphorus, potassium, calcium and magnesium in 'Nikkey' cucumbers grown in complete nutrient solution (CS) with the omission of one of the macronutrients at the time of the visualization of the first symptom of deficiency

Nutritive solution		Leaf content (g kg ⁻¹)				
		N	Р	K	Ca	Mg
CS	NL	61,3	6,9	21,5	6,3	3,3
	IL	57,8	5,3	28,5	11,7	2,9
	OL	53,8	6,3	23,2	34,7	5,2
Omission of the nutrient	NL	33,4	5,3	13,8	1,5	1,6
in the column	IL	18,9	1,5	9,5	3,1	1,2
	OL	20,5	2,0	14,1	19,1	2,4

(NL - new leaf; IL - intermediary leaf; OL - old leaf)

With a continuous omission of N, the chlorosis of intermediary leaves was stronger and older leaves began to show an initial yellowing of leaves, whereas leaves close to the meristem did not manifest symptoms. The chlorosis of older leaves is the typical initial symptom of N deficiency, because the plant redistributed it from older and intermediary leaves to organs or younger leaves [14] aiming to ensure growth and development, since it is a nutrient with an easy redistribution in the plant via phloem and due to the paucity of the nutrient in the medium.

There was a widespread chlorosis in intermediary and old leaves seven days after the omission. At this time, the plants had a growth lower than those grown in CS, consistent with the observations by [15] and [6] for cabbage and cucumber, respectively. The authors stated that the N deficiency limited the most the production of dry matter.

Ten days after the omission of N, there was a widespread chlorosis (Fig. 2), including new leaves that was also chlorotic. This is attributed to the high demand of N, because plants were at the 49th day of the cycle and bearing fruits [10]. Therefore, in this moment, in the absence of this nutrient in the medium, fruits became very strong drains over vegetative structures [16].



Fig. 2. Widespread chlorosis in 'Nikkey' cucumber plants after 15 days of N omission in nutrient solution

3.2 Phosphorus

The first P deficiency displayed on cucumber plants occurred at 20 days after treatment

initiation. A loss of green hue in intermediary leaves was observed, which had a tanned coloring compared to those grown in CS (Fig. 3). Just as observed for plants with N omission, plants grown with P omission redistributed the nutrient of intermediary and old leaves to leaves close to the meristem [7], explaining the green color loss of intermediary leaves and the low leaf content as a result of nutrient redistribution (Table 1).

At the visualization of the symptom, the P content in new, intermediary and old leaves under P omission were lower to those observed in the respective leaves of plants grown in CS (Table 1). Although the P content in old leaves (2.0 g kg⁻¹) was lower than what is considered adequate for cucumbers, i.e., 3 to 6 g kg⁻¹ [1], the loss of green color was not seen in these leaves.



Fig. 3. Browning of the intermediary leaf of 'Nikkey' cucumber as an effect of omitting P (right) compared to the complete solution (left)

Unlike what was observed for N omission, which quickly slowed growth even after omitting P, the cucumber plant grew normally. Upon visualizing the symptom, the plants had 23 knots and had 2 m length from the main stem, similar to plants grown in CS. [17] observed that the omission of P did not dramatically paralyze cucumber plant's growth. The result may be due to a small demand for the nutrient, mainly in a vegetative stage of the culture [10], and its high mobility in the plant [14], also explaining the delay in the initial manifestation of symptoms in this study.

With the evolution of P deficiency, a slight shriveling of the limbs of intermediary leaves was observed and, unlike the uniform yellowing caused by the deficiency of N, P omission caused chlorosis of randomly distributed areas in the leaf blade, like a mosaic, advancing from the petiole insertion region of the limbus to the edges (Fig. 4).

At the end of the experiment, after 27 days of P omission in the nutrient solution, chlorosis evolved to necrosis and burning of the edges, consistent with the findings of [18], who reported necrosis in mini-cucumber leaves deficient in P at the end of the experiment.

3.3 Potassium

Early symptoms of K deficiency were observed two days after the omission of K in the nutrient solution. They were characterized by chlorosis in the blade's apical edge of intermediary leaves from primary and secondary rods, progressing to the center of the leaf, in tissues located between the veins (Figs. 5A-B). Symptoms are those found by [15] and [17]. However, [17] observed that the symptoms started on older leaves. The explanation for observing K deficiency symptom

in intermediary leaves, different from that observed by [17], probably it was due to the K omission time, which in this case was in early fruiting. [17] omitted the nutrient at the beginning of vegetative stage. As the cucumber fruits are strong K drains, the demand for nutrient may have been provided by nearest leaves instead of the older leaves, therefore by intermediary leaves.

At the visualization of the deficiency, the K content in intermediary leaves was 9.5 g kg⁻¹, lower than the 16-30 g kg⁻¹ range considered appropriate to cucumber [1]. However, new and old leaves that did not show symptoms at this time were also deficient (Table 1).

Yellowing edges of intermediary leaves of primary and secondary stems were observed four days after the omission of K in the nutrient solution, which subsequently developed to necrotic tissue and the chlorosis moved to the center of the limbus (Fig. 5C), while in older leaves symptoms were not observed.



Fig. 4. Initial chlorosis in 'Nikkey' cucumber leaves as an effect of omitting P in the nutrient solution

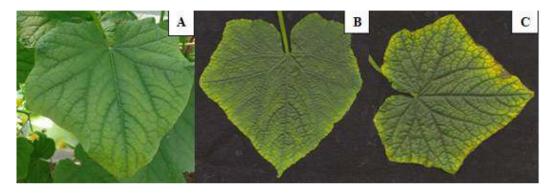


Fig. 5. Initial chlorosis (A - B) and advance to marginal necrosis (C) in intermediary leaves of 'Nikkey' cucumber as an effect of the omission of K in the nutrient solution

After nine days of omission of K in the nutrient solution, the plants had similar height, number of nodes and branches as those grown in CS. However, the necrosis of intermediary leaves advanced to the inside of laminas. The tissue necrosis in plants with K deficiency is attributed to the accumulation of putrescine, a polyamine that in high concentrations cause cellular imbalances and necrosis of plant tissues [19].

At the 59th day of the cycle, the leaves near the meristems began to manifest yellowing at the edges of leaves near apical regions, advancing to the center of leaf blades, similar to that previously observed with intermediary leaves. To the symptom is attributed the excessive synthesis of reactive oxygen species (ROS) that caused chlorophyll degradation and damage to cell membranes, which then causes chlorosis of leaf tissue [20,21].

3.4 Calcium

Ca deficiency symptoms in cucumber plants began three days after Ca omission in the nutrient solution, characterized by randomly scattered chlorotic stains in new leaf limbs (Fig. 6A). This symptom developed to necrotic points, which were observed in new and intermediary leaves located up to the sixth leaf node from the apical meristem of the main stem (Fig. 6B) five days after nutrient omission. This is attributed to cell disruption and to subsequent collapse of plant tissues, as observed by [17] with cucumber.

This quick view of symptoms is attributed to the involvement of Ca in structuring and cell

stabilization [1]. Therefore, among macronutrients, Ca is in greater proportion in the middle lamella of the cell wall as pectate, contributing to cell expansion and elongation [22].

Upon viewing the initial symptom of Ca deficiency, the nutrient content in new, intermediary and old leaves were 1.5, 3.1 and 19.1 g kg⁻¹ (Table 1). Therefore, only old leaves had Ca in an adequate concentration for the culture. However, when considering the range 13 to 35 g kg⁻¹ proposed as adequate for cucumber [1], both young and intermediate leaves of plants grown in CS would be with nutrient deficiency too. It is noteworthy that in the time of assessment of foliar content, only Ca deficiency symptoms were observed in young leaves of plants cultivated with the omission of this nutrient.

Both in Ca omission and CS, the concentration of nutrient increases in older parts of the plant (Table 1) due to its very low mobility [23,14] and low transpiration rate of the developing organs [8], which decreases the strength of these drains in the partition of Ca absorbed by plants.

Ten days after the omission of Ca, with the development of the deficiency, there was an intense necrosis of the tissue located in leaf edges, forming a tipburn, a characteristic symptom of Ca deficiency [8]. This physiological deficiency was predominantly observed in growing leaves [24,25] close to the top of stem, which were severely shriveled (Fig. 7).

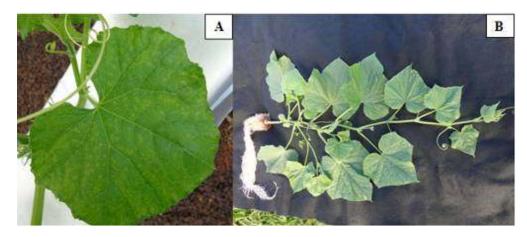


Fig. 6. Chlorotic stains scattered on the blade of a new leaf (A) and development of the deficiency (B) on 'Nikkey' cucumber as an effect of Ca omission in the nutrient solution



Fig. 7. Tipburn, shriveling and chlorosis in a new leaf of 'Nikkey' cucumber as an effect of Ca omission in the nutrient solution

[17] described poor leaf formation in a Ca deficient cucumber, with the margin of the leaves turned upwards, while [22] reported that plants with Ca deficiency do not have a normal morphogenesis of growing organs, since they demand high and continuous amounts of Ca to the stabilization of cell wall and plasma membrane integrity.

Fifteen days after the omission, when the plants were at the 54th day of the cycle, it was found that both the emission of flowers and the fruit growth decreased considerably compared to plants in CS. In addition, it was possible to observe necrotic tendrils and symptoms of apical rot (Fig. 8), a physiological deficiency of various fruit vegetables [26] caused by an inadequate concentration of Ca in the affected tissues [3,5].

3.5 Magnesium

Similar to what happened to N, P and K, known as mobile nutrients [7], the initial observation of Mg deficiency symptoms began in intermediary leaves, differing partly in the initial location of symptoms as reported in the literature [14].

The omission of nutrients in a high nutritional demand stage [10] may explain this difference, because, with the abrupt withdrawal of nutrients from nutrient solutions during the period when plants started fruiting, they were redistributed to areas closer to the growing regions, i.e., from intermediary leaves to new leaves. In this context, the sudden interruption of nutrients (omission) may lead to different responses from those of field conditions (deficiency), where nutrient availability is less variable [8].

The early symptom of Mg deficiency was observed five days after its omission from the nutrient solution and it was characterized by a slight chlorosis among secondary veins of intermediary leaves (Fig. 9).

At this time, the plants in Mg omission had 1.6, 1.2 and 2.4 g kg⁻¹ in new, intermediary and older leaves, respectively. These contents are lower than those considered suitable by [1], i.e., 3-6 g kg⁻¹. Thus, both young and intermediary leaves in CS were also deficient (Table 1).

Similar to what happened with K deficiency, the chlorosis motivated by -Mg is explained by the excessive accumulation of reactive oxygen species (ROS), which causes severe oxidative damage to thylakoids and hence the breakdown of chloroplasts [27].

Leaving it in the solution without Mg, interveinal chlorotic areas became whitish and the shriveling of the leaf edges was observed (Fig. 9). In advanced stages of Mg deficiency, the chlorotic lesions among veins advance to the disintegration of the mesophyll. Therefore, only the unchanged external cuticle remains, causing the whitish appearance of leaves [17].

Severe deficiency symptoms were observed 16 days after Mg omission in older leaves, while it was not perceived in those next to meristems. During this period, the plants had developing fruits, but with symptoms of collapse of apical tissues, similar to what happened in the deficiency of Ca.



Fig. 8. Apical rot in fruits of 'Nikkey' cucumber as an effect of Ca omission in the nutrient solution

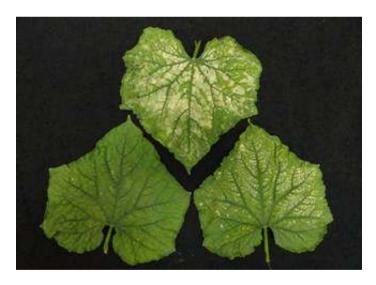


Fig. 9. Magnesium deficiency symptom evolution in an intermediary leaf of 'Nikkey' cucumber

4. CONCLUSION

With the omission of nutrients in early fruiting plants, the symptoms of the deficiencies of N, P, K and Mg are observed initially in intermediary leaves and Ca deficiency in young leaves.

The deficiency of N initially causes uniform chlorosis in the leaf blade of intermediary leaves, a symptom that advances to all leaves and causes strong constraints on the growth of cucumber plants.

With the omission of P, symptoms begin with the loss of the characteristic green color of cucumber plants in the blade of intermediary leaves, evolving to chlorotic areas randomly distributed both in intermediary and in old leaves, and it culminates in tissue necrosis.

The omission of K causes chlorosis symptoms in intermediary leaves, beginning from the apical margin and progressing to the entire limb, culminating in necrosis.

The Ca omission initially causes chlorotic stains randomly scattered over young leaves of limbs. They advance to intermediary leaves with tissue necrosis, finally culminating in apical rot in fruits.

With the omission of Mg, symptoms begin with internerval chlorosis of intermediary leaves, evolving to whitish color and necrotic tissue.

The contents of N, P, K, Ca and Mg in leaves, according to the visualization of deficiency symptoms, are 18.9, 1.5, 9.5, 1.5 and 1.2 g kg⁻¹, respectively.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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