Impact of nanofertilizers on cucumber yield and quality under greenhouse conditions

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Abstract
Nanofertilizers represent an efficient method for fertilizing crops, with a positive impact on plant development. This work aimed to determine the impact of Nubiotek Ultra Ca and Nubiotek Hyper Fe +Mg nanofertilizers on the yield and quality of cucumber fruits in 2020. Two doses of Nubiotek Ultra Ca (0 and 20 L ha⁻¹) and four doses of Nubiotek Hyper Fe+Mg (0, 2, 4, and 8 ml L⁻¹) were evaluated. An experimental design of randomized complete blocks was used with a factorial arrangement (2x4), with four repetitions. The results showed that the nanofertilizers evaluated did not improve internode distance, dry weight of each organ, root length, total soluble solids, vitamin C, titratable acidity, and fruit color parameters compared to the values obtained with the control. On the other hand, with the treatments of 0 L ha⁻¹ Nubiotek Ultra Ca + 8 ml L⁻¹ Nubiotek Hyper Fe+Mg and 20 L ha⁻¹ Nubiotek Ultra Ca + 2 ml L⁻¹ Nubiotek Hyper Fe+Mg, the fruit yield increased by 111 and 123%, respectively, compared to the control. Likewise, a greater firmness of fruits was observed when applying Nubiotek Ultra Ca and Nubiotek Hyper Fe+Mg in doses of 20 L ha⁻¹ + 8 ml L⁻¹; on the other hand, the highest content of chlorophyll a, b and total was achieved with doses of 20 L ha⁻¹ + 4 ml L⁻¹ and 20 L ha⁻¹ + 8 ml L⁻¹.

Keywords:
Cucumis sativus L., chlorophyll, firmness.
Introduction

Cucumber (*Cucumis sativus* L.) is the second most widely cultivated cucurbit in the world (*Singh et al., 2020*). Nonetheless, cucumber production requires moderate to high nutrient rates to achieve better yield and higher quality (*Kumar, 2020*). Fertilizers are known to be vital agricultural inputs that improve crop production by 30 to 50% (*Chen and Yada, 2011*).

Nevertheless, due to the low efficiency of conventional fertilizers, less than half of the amount applied is used efficiently by plants (*Congreves et al., 2021*), the rest seeps into the soil or is not available for crops, causing groundwater contamination and reduced soil fertility (*Verma et al., 2022*). Therefore, it is necessary to use efficient alternatives in crop nutrition to solve the problems caused by the application of high rates of conventional fertilizers and, at the same time, increase food production.

Nanofertilizers represent an alternative to improve the efficiency in the use of nutrients, reduce the use of conventional fertilizers, consequently, reduce adverse impacts on the environment. Nanofertilizers are nutrient compounds of nanostructured formulations (*Raliya et al., 2017; Astaneh et al., 2021; Rajput et al., 2021*).

The beneficial effect of nanofertilizers on crop development, compared to conventional fertilizers, is due to the fact that they have a particle size of 1-100 nm and a larger specific surface area, which means that they have a higher dissolution rate and are easily absorbed through the leaves or roots (*Verma et al., 2022*); in addition, the release of nutrients from nanoparticles is done at a slow rate, thus allowing for a prolonged duration of effective nutrient supply for plants (*Verma et al., 2022*).

Recent advances in sustainable agriculture have seen the beneficial use of various nanofertilizers to increase crop production. As reported by *Rajput et al. (2021)*, more than 102 nano-enabled fertilizers are available on the market in 17 countries and in Mexico there are two enabled lines, Nubiotek Ultra and Nubiotek Hyper (*Menossi et al., 2022*).

However, there is little research indicating the influence of nanofertilizer fertilization on vegetable development and, above all, there is no research on the effect of nano-enabled fertilizers available in Mexico on crop production. Therefore, in this research work, the objective was to determine the impact of the Nubiotek Ultra Ca and Nubiotek Hyper Fe+Mg nanofertilizers on the yield and quality of cucumber fruits.

Materials and methods

Description of the study area

The research work was carried out in 2020 in a chapel-type greenhouse with a polycarbonate cover at the Antonio Narro Autonomous Agrarian University in Saltillo, Coahuila, Mexico. Seeds of cucumber cv. Centauro were used and were sown in 8 L black polyethylene containers, using a mixture of acid peat moss and perlite (1:1%, v:v) as a growth medium. In order to have uniform plants, sowing was done directly, placing one seed per container at a distance of 40 cm between plants and 80 cm between rows.

Description of nanofertilizers and treatments

The nanofertilizers used are the commercial products Nubiotek Ultra Ca and Nubiotek Hyper Fe +Mg. Nubiotek Ultra Ca contains 13% Ca, 17% N, 3.5% K and 17% humic acids, while Nubiotek Hyper Fe+Mg provides 1.5% Fe and 0.5% Mg. The treatments used were two doses of Nubiotek Ultra Ca (0 and 20 L ha\(^{-1}\)) and four doses of Nubiotek Hyper Fe+Mg (0, 2, 4, and 8 ml L\(^{-1}\)), making a total of eight treatments.

In all treatments, the nutrient solution proposed by *Steiner (1961)* (meq L\(^{-1}\): 12 NO\(_3\), 1 H\(_2\)PO\(_4\), 7 K, 4 Mg, 9 Ca, and 7 SO\(_4\)) and the following micronutrients (mg L\(^{-1}\)): 6 Fe, 0.48 Zn, 2.96 Mn, 0.32 B, and 0.24 Cu, were used as fertilization base. For the formulation of the base nutrient solution, the
chemical properties of the irrigation water were considered. The pH of the solution was adjusted to 6 ±0.1 before each irrigation with 1 N H\textsubscript{2}SO\textsubscript{4}. Application of the Steiner solution begun 15 days after emergence (DAE).

Irrigations were carried out manually according to the water needs of the plants, applying a sufficient volume of the nutrient solution to maintain a leachate fraction of 20%. The treatments were applied every 15 days, starting at 21 DAE. Treatments with Nubiotek Ultra Ca were added towards the base of the stem of the cucumber plants and doses of Nubiotek Hyper Fe+Mg were applied foliarly.

**Variables evaluated**

The experiment ended 100 days after sowing, the fruit harvest was carried out when they presented the characteristic size of the variety. The weight of each fruit (g) was evaluated with the help of a digital scale and the yield per plant was determined by summing the total weight of harvested fruits. In addition, the content of total soluble solids, vitamin C, titratable acidity, firmness, chlorophyll (a, b and total), and color parameters (L\textsuperscript{*}, a\textsuperscript{*} and b\textsuperscript{*}) of the epidermis of the fruits were obtained.

The total soluble solids content was assessed by placing a drop of fresh juice in the prism of a digital refractometer (Atago\textsuperscript{®}, USA Inc., Bellevue, WA, USA). The vitamin C content of the fruit was determined by the titration method with 2,6-dichlorophenolindophenol (Padayatt et al., 2001).

Titratable acidity was determined according to the method of the (AOAC, 2000). A sample of 10 ml of fruit juice was taken and adjusted to 125 ml with distilled water. This solution was titrated with 0.01N NaOH to a pH of 8.3. The results of these measurements were expressed as a percentage of citric acid by applying the following formula:

\[
\frac{V_{NaOH} \times N_{NaOH} \times meq_{acido} \times 100}{V}
\]

Where: \(V_{NaOH}\) = volume of NaOH used for titration; \(N_{NaOH}\) = NaOH normality; \(Meq_{acido}\) = acid milliequivalents; \(V\) = ml of the sample. The base-to-acid equivalent value for citric acid is: 0.064.

Firmness was determined at three points of the fruit using a PCE-PTR 200 digital penetrometer, equipped with an 8 mm diameter convex tip. The chlorophyll a, b, and total content of the epidermis of the fruit was quantified by spectrophotometry (Wellburn, 1994). The color parameters a\textsuperscript{*}, b\textsuperscript{*} and L\textsuperscript{*} of the epidermis of the fruit were measured on two opposite sides of the equatorial part using a Minolta Chroma MeterCR-400 equipment (MinoltaCorp, Ramsey, New Jersey, USA).

At the end of the experiment, the internode distance of the plants was evaluated, then the roots were washed with drinking water and distilled water to remove excess substrate. The plant was separated into roots, stems, and leaves to determine root length. The separated organs were placed in a drying oven at 65 °C for 72 h, after which the weight of the dry matter was recorded with an analytical balance.

**Experiment design and data analysis**

The work was carried out under a randomized complete block design with a 2x4 factorial arrangement, with four repetitions per treatment. The data obtained were subjected to an analysis of variance and comparison of means with Tukey’s test (\(\alpha\leq 0.05\)) using the SAS statistical program version 9.2 (Statistical Analysis Systems).

**Results and discussion**

In this work, it was observed that the Nubiotek Ultra Ca nanofertilizer induced significant changes (\(\rho\leq 0.05\)) on internode length, stem dry weight and yield; likewise, the Nubiotek Hyper Fe+Mg nanofertilizer significantly affected (\(\rho\leq 0.05\)) leaf dry weight, root length and yield, without observing a positive effect of these nanofertilizers on the growth of cucumber plants; on the other hand, the interaction of the two nanofertilizers evaluated statistically affected (\(\rho\leq 0.05\)) root length and fruit yield (Table 1).
Table 1. Effect of nanofertilizer doses on growth and yield parameters in cucumber plants.

<table>
<thead>
<tr>
<th></th>
<th>Internode length (cm)</th>
<th>Dry weight (g plant⁻¹)</th>
<th>Root length (cm)</th>
<th>Yield (g plant⁻¹)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Leaf</td>
<td>Stem</td>
<td>Root</td>
</tr>
<tr>
<td>Nubiotek Ultra Ca (L ha⁻¹)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0</td>
<td>14.63a</td>
<td>36.58a</td>
<td>17.44a</td>
<td>2.69a</td>
</tr>
<tr>
<td>20</td>
<td>13.95b</td>
<td>32.67a</td>
<td>14.87b</td>
<td>2.19a</td>
</tr>
<tr>
<td>Nubiotek Hyper Fe+Mg (ml L⁻¹)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0</td>
<td>14.57a</td>
<td>36.17ab</td>
<td>16.62a</td>
<td>2.25a</td>
</tr>
<tr>
<td>2</td>
<td>14.14a</td>
<td>26.83c</td>
<td>13.87a</td>
<td>2.5a</td>
</tr>
<tr>
<td>4</td>
<td>14.32a</td>
<td>33bc</td>
<td>17.37a</td>
<td>2.5a</td>
</tr>
<tr>
<td>8</td>
<td>14.15a</td>
<td>42.5a</td>
<td>16.75a</td>
<td>2.5a</td>
</tr>
</tbody>
</table>

Anova (p#0.05)

- Nubiotek Ultra Ca: 0.045 0.06 0.047 0.076 0.062 0.009
- Nubiotek Hyper Fe+Mg: 0.763 0.001 0.212 0.883 0.001 0.006
- Interaction: 0.159 0.674 0.572 0.35 0.006 0.005

Means with different letters indicate significant differences according to Tukey’s test at α≤ 0.05.

According to the interaction of the two nanofertilizers evaluated, it was observed that, when applying the Nubiotek Hyper Fe+Mg nanofertilizer, alone or in combination with the Nubiotek Ultra Ca nanofertilizer, the root length of the cucumber plants was not improved, in fact, when applying the treatments 0 L ha⁻¹ Nubiotek Ultra Ca + 2 ml L⁻¹ Nubiotek Hyper Fe+Mg, 0 L ha⁻¹ Nubiotek Ultra Ca + 4 ml L⁻¹ Nubiotek Hyper Fe+Mg, 20 L ha⁻¹ Nubiotek Ultra Ca + 2 ml L⁻¹ Nubiotek Hyper Fe+Mg and 20 L ha⁻¹ Nubiotek Ultra Ca + 4 ml L⁻¹ Nubiotek Hyper Fe+Mg, a shorter root length was obtained, compared to the control, decreasing by 19.3, 19.3, 31.1, and 16.2%, respectively (Figure 1A).
Figure 1. Effect of the interaction of nanofertilizer doses on root length (A); and yield (B) of cucumber. Means with different letters indicate significant differences according to Tukey’s test at $\alpha \leq 0.05$. The bars indicate the standard error of the mean (n= 4).

This is not consistent with what was reported by Kanwar et al. (2019), who have indicated that plants exposed to nanofertilizers present vital changes in biological and physiological processes, inducing improvements in crop growth. Verma et al. (2022) noted that the effects of nanofertilizers on plant development may be regulated by soil characteristics, the environment, the delivery mechanism of nanofertilizers, and plant species.

Contrary to what was observed in the behavior of root growth when applying nanofertilizers, the yield of cucumber fruit was higher than the control when adding the treatments 0 L ha$^{-1}$ Nubiotek Ultra Ca + 8 ml L$^{-1}$ Nubiotek Hyper Fe+Mg and 20 L ha$^{-1}$ Nubiotek Ultra Ca + 2 ml L$^{-1}$ Nubiotek Hyper Fe+Mg, exceeding it by 111 and 123%, respectively (Figure 1B).

These results are consistent with those reported by Rahman et al. (2021), who noted that total tomato yield increased by 31.8% in plants in which a mixture of nanofertilizers (Zn, Fe and Cu) was added, compared to the control, as a result of the balanced and controlled distribution of nanofertilizers within the plant. Authors such as Morsy et al. (2018), when evaluating a nanofertilizer that provides Ca, Mg and Fe in wheat (Triticum durum L.) crops, reported a higher yield, attributing this effect to the efficiency of nutrient use and improved photosynthesis by the plant.

Likewise, the Nubiotek Ultra Ca nanofertilizer significantly affected ($p < 0.05$) the content of total soluble solids, vitamin C and fruit firmness, while the Nubiotek Hyper Fe+Mg nanofertilizer statistically affected ($p < 0.05$) the titratable acidity and fruit firmness; likewise, the interaction of the two nanofertilizers evaluated statistically affected ($p > 0.05$) total soluble solids, vitamin C, titratable acidity, and fruit firmness (Table 2).

<table>
<thead>
<tr>
<th>Nubiotek Ultra Ca (L ha$^{-1}$)</th>
<th>TSS (°Brix)</th>
<th>Vitamin C (mg 100 g$^{-1}$ FW)</th>
<th>Titratable acidity (% CA)</th>
<th>Firmness (N)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>2.59a</td>
<td>16.55b</td>
<td>0.125a</td>
<td>50.77b</td>
</tr>
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</table>
According to the interaction of the two nanofertilizers evaluated, it was observed that, although the treatments presented statistical differences in the parameters of total soluble solids, vitamin C and titratable acidity, none of the treatments in which the nanofertilizers were added exceeded the control (Figure 2 A, 2B and 2C). The results of this research are in line with the findings of Cvelbar et al. (2021), who observed that, when fertilizing strawberry (Fragaria x ananassa Duch.) plants with a nanofertilizer that provides Ca, Mg and Fe, the total soluble solids did not show an increase.
It has been pointed out that fruit quality parameters are mainly affected by nutrient availability during crop development (Li et al., 2021). Nanofertilizers improve the diffusion, solubility, and availability of nutrients for plants due to their small size, high specific surface area, and the synergism that nanoparticles present over conventional fertilizers (Verma et al., 2022).

According to the above, an improvement in the quality parameters of the cucumber fruits could be expected when adding the nanofertilizers, although this positive effect of the nanofertilizers was only observed in the firmness of the fruit since a better firmness of the fruit was obtained by applying 20 L ha⁻¹ Nubiotek Ultra Ca + 8 ml L⁻¹ Nubiotek Hyper Fe+Mg, exceeding the control by 18% (Figure 2D).

The above answer represents an advantage for a better consumer acceptance since fruit firmness is considered an attribute of great importance in cucumber crops, since the consumer looks for firm and crunchy fruits (Azarmi et al., 2015). Ca maintains the integrity and stability of the plasma membrane and cell wall through bonds established with proteins, phospholipids, and pectic acid (Langer et al., 2019; Sajid et al., 2020).
Nevertheless, Ca is a low-rate long-distance translocation ion in the xylem (Maathuis and Diatloff, 2013). Nubiotek Ultra Ca is considered an enantiomorph amphiphilic colloid, so it allows Ca to penetrate quickly and effectively into the interior of the plant; in addition, it allows it to be transported to all the organs of the plant. This quality of the Nubiotek Ultra Ca nanofertilizer may have been partly responsible for improving the firmness of cucumber fruits.

Similarly, in this work it was observed that the Nubiotek Ultra Ca nanofertilizer significantly affected (p≤ 0.05) the content of chlorophyll a, chlorophyll b, and total chlorophyll in the epidermis of cucumber fruits; likewise, Nubiotek Hyper Fe+Mg statistically influenced (p≤ 0.05) the content of chlorophyll a, chlorophyll b, total chlorophyll and L*, whereas the interaction of the two nanofertilizers evaluated statistically affected (p≤ 0.05) chlorophyll a, chlorophyll b, and total chlorophyll (Table 3).

<table>
<thead>
<tr>
<th>Chlorophyll a</th>
<th>Chlorophyll b</th>
<th>Total chlorophyll</th>
<th>L*</th>
<th>a*</th>
<th>b*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nubiotek Ultra Ca (L ha⁻¹)</td>
<td></td>
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<tr>
<td>0</td>
<td>0.037b</td>
<td>0.016b</td>
<td>0.053b</td>
<td>30.64a</td>
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<tr>
<td>20</td>
<td>0.047a</td>
<td>0.019a</td>
<td>0.068a</td>
<td>29.67a</td>
<td>-6.84</td>
</tr>
<tr>
<td>Nubiotek Hyper Fe+Mg (ml L⁻¹)</td>
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<td></td>
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<tr>
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<td>0.169</td>
<td>0.08</td>
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</tr>
</tbody>
</table>

Means with different letters indicate significant differences according to Tukey’s test at α≤ 0.05. FW= fresh weight.

The interaction of the two nanofertilizers showed that applying 20 L ha⁻¹ Nubiotek Ultra Ca + 4 ml L⁻¹ Nubiotek Hyper Fe+Mg and 20 L ha⁻¹ Nubiotek Ultra Ca + 8 mL L⁻¹ Nubiotek Hyper Fe+Mg resulted in the highest content of chlorophyll a, b and total, surpassing the control (Figure 3A, 3B and 3C). In relation to the above, it has been indicated that nanofertilizers cause an increase in chlorophyll content because they induce an increase in physiological and biochemical indices in plants (Pirvulescua et al., 2015).
In addition to the above, N and Mg, being nutrients that are part of the structure of chlorophyll (Khalil et al., 2021; Mohammed et al., 2021), by increasing the rate of N and Mg through the addition of Nubiotek Ultra Ca and Nubiotek Hyper Fe+Mg nanofertilizers, were able to cause an increase in chlorophyll content in cucumber fruits. Likewise, the additional application of Fe by Nubiotek Hyper Fe+Mg and its easy absorption could cause a higher chlorophyll content in cucumber fruits, since it has been indicated that Fe is essential in the biosynthesis of the chlorophyll molecule within plants (Hamouda et al., 2016).

Conclusions

In this work, it was found that the Nubiotek Ultra Ca and Nubiotek Hyper Fe+Mg nanofertilizers increase the yield of cucumber fruits, mainly when both nanofertilizers are added in doses of 0 L ha\(^{-1}\) + 8 ml L\(^{-1}\) and 20 L ha\(^{-1}\) + 2 ml L\(^{-1}\). In addition, applying Nubiotek Ultra Ca and Nubiotek Hyper Fe+Mg improves the firmness and chlorophyll content of cucumber fruits.

Bibliography


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