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IMPACT OF UREA NUTRITION ON SOIL AND PLANT CHEMICAL VARIABLES AND DEVELOPMENT OF MEXICAN LIME

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Abstract: The yield of Mexican lime in Michoacan, Mexico, is notably variable, suggesting possible nutritional issues. Nitrogen (N) fertilization is key to reaching the crop's potential; however, various fertilizer sources are used without considering their suitability for local soils, leading to variable effectiveness. Moreover, different fertilization strategies interfere, as they often do not reflect in plant quality. The objective was to determine the effect of urea doses (46% N) and its collateral impact on soil/plant chemical variables. A study was conducted on Mexican lime, where urea doses (41.66, 83.33, 125, and 0 g per plant in 12 applications every 15 days) were evaluated, to determine their effects on different edaphic and vegetative variables. Soil pH, electrical conductivity (E. C.), and N and potassium (K) levels were analyzed, as well as leaf N and K levels, bud number and length, and yield. Results showed statistical differences in soil pH, E. C., N, and K levels, as well as in leaf N and K levels. The set of treatments with urea was clearly distinguished from the control. Plant development characteristics were also statistically different, due to urea application. The highest observed yield was 112.5 kg per plant, corresponding to the third treatment, which received 125 g of urea per plant.

Keywords: *Citrus aurantifolia*, soil electrical conductivity (E. C.), nitrogen (N), soil pH, potassium (K).

INTRODUCTION

According to FAO, in 2021, Mexico ranked second worldwide in citrus harvested area, with 195,619 ha, only behind India, which reported 327,000 ha. In terms of production, Mexico reached 2,983,802 tons, while India produced 3,548,000 tons. However, in terms of yield, Mexico ranked 39th with 15.25 tons·ha^{-1} (FAOSTAT, 2023). Regarding *Citrus aurantifolia* (Christm.) Swingle (Mexican lime), Mexico is considered the world's leading producer, with a national distribution reported in 28 states covering 199,650 ha and 3,071,969.97 tons of production in 2022. Particularly, the state of Michoacan led production with 54,157 ha harvested and 856,738 tons produced, distributed across 32 municipalities (SIAP-SADER, 2023).

In relation to the management of Mexican lime plantations in Michoacan, several agricultural practices, when poorly implemented or deficient, result in low production, and fruit quality is affected (Martínez *et al.*, 2023). Notable deficiencies include: pest control management based on prevention is uncommon, since attention is mostly reactive or curative; the use of unadapted rootstocks, which indirectly depends on the producer, as the plants are sourced from commercial nurseries that do not certify that the acquired material corresponds to the genotypes, both rootstocks and scions, suited to the edaphic-environmental conditions of the area; low population densities, as wide planting frames still prevail, and little progress has been made in testing new intensive production systems; the excessive and unbalanced induction of flowering, as flower induction is often confused with severe pruning, which shortens the tree's juvenile phase, accelerates its aging, and deteriorates plant vigor; improper irrigation management, as there is no clear understanding of how long and how frequently irrigation should be applied, much less based on soil physical variables and their interpretations concerning critical moisture levels; the lack of soil conditioners in the clay soils prevalent in the region, which requires chemical evaluation, through soil analysis to determine the most effective amendments; and unbalanced nutrition, where various criteria are used, but the production goal is often not considered important and is based on common sense. This last aspect has a notably marked effect, as the average yield of Mexican lime is around 15 tons·ha^{-1}, yet the species has the potential to produce up to 45 tons·ha^{-1}. This discrepancy suggests the possible existence of nutritional issues (Maldonado *et al.*, 2001).

As mentioned, after water and temperature, fertilizers are considered the third most important factor in agricultural crop production (Morales-Morales *et al.*, 2019). Nitrogen is the element most required by plants due to its marked influence on growth and production, as it is part of proteins, chlorophyll, and other metabolic processes. Citrus requires nitrogen in greater proportions; therefore, the range of commonly used nitrogen fertilizers includes ammonium sulfate $(NH_4)_2SO_4$, urea $CO(NH_2)_2$, ammonium nitrate NH_4NO_3, diammonium phosphate $(NH_4)_2HPO_4$, and monoammonium phosphate $NH_4H_2PO_4$. In the lime-producing region of the Apatzingan Valley, the vertisol soils tend to be clayey with alkaline pH, so urea can be continuously used as a nitrogen fertilizer source. Importantly, urea has an acidic reaction, affecting pH and triggering positive collateral effects on other chemical variables. Additionally, it provides a high nitrogen content (46%). Its chemical formula is $CO(NH_2)_2$, which, when hydrolyzed, produces ammonium and bicarbonate. Although bicarbonate ions react with pH, this is only temporary, and the reaction quickly reverses as ammonium releases hydrogen, entering the nitrification process with the involvement of specialized bacteria. This means that ammonium ions (NH_4+) are oxidized to nitrite (NO_2-), and nitrite is quickly oxidized to nitrate (NO_3-). Urea decomposition in soil occurs within an average period of four to seven days. During this process, both ammonium and nitrate are forms that can be absorbed by plants (Morales-Morales *et al.*, 2019). With urea as a nitrogen fertilizer source for Mexican lime, the leaf area is noticeably increased, leaves thicken, photosynthesis is more active, and fruit yield and quality improve (Morales--Morales *et al.*, 2019). It is important to note, that the chemical reactions that occur in the soil due to nitrogen fertilizer supplies like urea, can alter the response of other elements, particularly potassium. In Mexican lime, potassium is the element absorbed in greater amounts than any other nutrient. Once potassium enters the vascular system through the plant's root, it is transported to various organs, as it is highly mobile, moving both up and down through the xylem and phloem. Potassium generally ranges from 1 to 6% (10,000 to 60,000 ppm) of the dry matter content (Leigh and Wyn-Jones, 1984).

Therefore, nutrient monitoring through soil chemical analysis is considered essential to determine fertilization needs and microelement deficiencies. Similarly, foliar analysis is useful for detecting nutritional deficiencies or excesses in the plant. It is recommended to perform this analysis at least once a year to optimize fertilizer use. With this support and the continuous observation of orchard management factors, producers can adjust nutrient doses in their fertilization programs (Maldonado *et al.*, 2008). Based on the aforementioned and to understand the interactive soil/plant dynamics of nitrogen sourced from urea fertilizer, the objective was to determine the effect of urea doses (46% N) and its collateral impact on soil/plant chemical variables.

MATERIALS AND METHODS

The experimental work was carried out in a plot established with Mexican lime trees in the production stage and grafted onto (*Citrus macrophylla* Wester, aka Alemow), located in the ejido Antunez, Michoacan, Mexico. The predominant climate in the area is classified as Bs_1, belonging to the group of dry climates; the least dry of the BS types, very hot, with an average annual temperature > 22 °C, and the coldest month's temperature > 18 °C. The rainfall regime occurs in summer, with at least 10 times more rainfall in the wettest month of the warm half of the year than in the driest month, and winter rainfall accounting for < 5% of the annual total, with little variation (Köppen modified by García, 2004). The region supports species of low deciduous forest, and the soil is classified as pelic Vertisol (INEGI, 2016).

The general plant management followed the recommendations issued by the National Institute of Forestry, Agriculture, and Livestock Research (Coria *et al.*, 2017). During the fall-winter season, the trees were pruned to remove unproductive and dry branches. The soil was tilled through mechanical harrowing to remove weeds and facilitate the placement of fertilizer around the drip zone, following the perimeter of the tree's canopy, encircling the moisture bulb. The soil was classified as pelic vertisol with clay texture, pH of 7.72, electrical conductivity of 0.33 dS/m, N levels of 5.83 $mg \cdot kg^{-1}$, and K levels of 779 $mg \cdot kg^{-1}$.

The experiment was designed as a randomized complete block with four treatments and four replicates. Each treatment consisted of two plants, totaling 32 selected plants. The treatments consisted of the addition of urea fertilizer (46% nitrogen) applied every 15 days to each plant over 12 applications: T1) 41.66 g; T2) 83.33 g; T3) 125 g; T4) 0 g (control).

Data were collected at varied intervals over 180 days for the following soil chemical variables: pH, electrical conductivity (E. C.), nitrogen (N), and potassium (K). In the plants, N and K levels were recorded, as well as the number of buds per branch per plant, bud length, and yield per plant. Soil sampling involved monthly collection per treatment/ replicate. The soil samples were prepared as wet paste, and a suction tube extractor was used to obtain the liquid solution. A manual potentiometer was used to measure pH, a conductimeter for E. C., and manual ionometers for N and K. For leaf N and K measurements, 10 healthy, fully developed leaves, along with petioles, located in the middle of a fruitless branch, were collected. The plant tissue was placed in a "Ziploc" plastic bag for grinding, and at least one drop of cell juice was extracted to be placed on the ionometer electrode, where readings were recorded. It is important to note that the data obtained from each sampling during the experimental period were averaged for each variable, except for bud length, which was analyzed both as an average and as a cumulative value. Variance analysis and means comparison using Tukey's test ($P = 0.05$) were performed on all variables using SAS statistical software version 9.2 (2019).

RESULTS

The response of Mexican lime to nitrogen doses and control treatments on the soil variables pH and electrical conductivity is presented in Figure 1. As observed, the analysis of variance showed significant statistical differences. For the pH variable, treatment 4 (control), which did not receive N, exhibited its natural pH level of 7.82 and, along with treatment 1 (low dose), was statistically superior. Treatments 2 and 3 (medium and high doses, respectively) showed lower pH values, with reductions of 6% and 7%, respectively, compared to the control, due to the effect of nitrogen. For the E. C. variable, the behavior was the opposite of that observed for pH. Taking the lowest value (control) as a reference, treatment 3 (high dose) showed a 46% increase, followed by treatment 2 (medium dose) with a 40% increase, and treatment 1 (low dose) with a 25% increase (Figure 1).

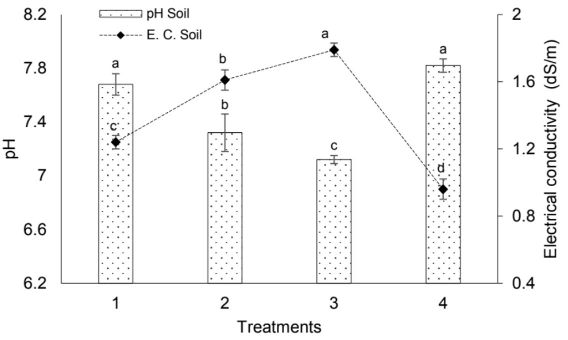

Figure 1. Analysis of variance for soil pH and E. C. (ds/m) variables and their response to urea addition in Mexican lime.

Regarding the chemical variables nitrogen and potassium in the soil and leaves, the analysis of variance revealed significant statistical differences (Figure 2). For the nitrogen variable in the soil, treatment 3 had the highest nitrogen content, followed by treatments 2 and 1, with the control being significantly surpassed by these treatments by 57%, 54%, and 40%, respectively. Similarly, for the nitrogen content in leaves, the trend was comparable to that of soil nitrogen, although it is noteworthy that in treatment 1, the nitrogen content in leaves was 26% higher than in the soil; in treatment 2, leaves nitrogen was 19% higher than soil nitrogen; and in treatment 1, leaves nitrogen exceeded soil nitrogen by 2% (Figure 2).

For the potassium variable in soil, statistical differences were present, but the values were close across the four treatments. However, for the potassium content in leaves, a notable inductive response was observed due to nitrogen and the plant's physiology, concentrating potassium in its tissues. Treatment 3, which had the highest nitrogen levels in both soil and leaves, also had the highest potassium concentration in leaves, followed by treatments 2 and 1. It is important to highlight that in the soil, across all treatments, potassium concentration in leaves increased by 93%, 90%, and 89% in treatments 3, 2, and 1, respectively. Even in the control, the potassium concentration increased by 87% (Figure 2). This response could be attributed to the ability of this plant species to effectively synthesize potassium, potentially drawing from existing soil reserves, influenced by the balanced cation/anion mass flow.

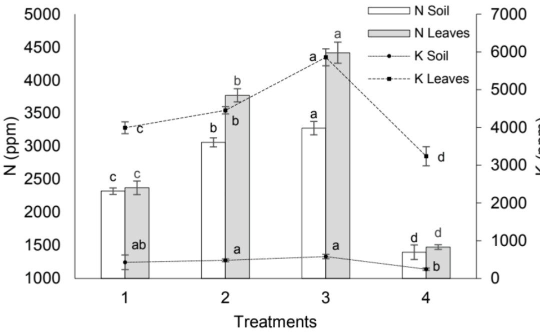

Figure 2. Analysis of variance for soil and plant N and K variables and their response to urea addition in Mexican lime.

The analysis of variance for development and yield variables is presented in Table 1. As shown, the analysis of variance revealed statistically significant differences. For the number of buds, the average bud length, cumulative bud length, and yield per plant, treatment 3 showed the highest values, followed by treatments 2 and 1, in order of nitrogen doses from highest to lowest (Table 1). In terms of the number of buds, treatments 3, 2, and 1 increased by 20%, 23%, and 7%, respectively, compared to the control. For bud length, treatments with nitrogen showed increases of 45%, 29%, and 26% for treatments 3, 2, and 1, respectively, compared to the control. Regarding yield per plant, treatment 3 surpassed the others by 16% over treatment 2, 30% over treatment 1, and 49% over the control (Table 1). These results clearly demonstrate the plant's response to nitrogen application, particularly to urea fertilizer under different dosing regimes.

Treatments	Number of buds[†]	Average bud length (cm)	Cumulative bud length (cm)	Yield/plant (kg)
1	14.71 ± 0.80 b	15.25 ±0.64bc	106.75 ± 4.5 bc	78.5 ± 5.97 c
2	15.61 ±1.79ab	18.16 ± 2.32 b	127.13 ± 16.23 b	94.25 ± 6.99 b
3	17.06 ± 0.78 a	23.37 ± 2.23 a	163.63 ± 15.71 a	112.5 ± 8.35 a
4	13.64 ± 0.25 b	12.84 ± 2.41 c	89.88 ± 16.87 c	57.5 ± 6.03 d
C. V.	6.95	11.70	11.71	8.05
P	0.004	0.0001	0.0001	0.0001

Table 1. Analysis of variance for development and yield variables and their response to urea addition in Mexican lime.

C.V. = Coefficient of variation; [†]Means ± standard deviation with different letters indicate differences, Tukey ($P \leq 0.05$).

The obtained results reflect the instability of nitrogen and potassium elements, as they are influenced by the tree's constant physiological activity. It should be noted that nitrogen is highly mobile within the plant, which explains its variable concentration in leaf tissues. Likewise, potassium is mobile within the plant due to the physiological activity of the fruit.

DISCUSSION

One of the factors that significantly affects the yield and quality of Mexican lime is fertilization, as adequate applications of nutrients, especially nitrogen, are required to achieve a good harvest (Perez-Zamora and Orozco-Romero, 2004). However, the losses of this element due to ionic interactions with soil conditions, application techniques, and irrigation management are considerable (Larios-González *et al*., 2021). In clay soils, characteristics such as the presence of free calcium carbonates and alkaline pH negatively affect the availability, accessibility, absorption, transport, and assimilation of various nutrients. In this regard, the use of soil amendments mitigates this problem (Pérez, 2002); however, they are not prioritized as a practice in Mexican lime cultivation.

It is noteworthy that nitrogen fertilizers, particularly urea, tend to acidify the soil and affect other variables (Boccolini *et al*., 2016). This was reflected in the present study, as treatments 1, 2, and 3, which received urea, averaged a 6% reduction in pH compared to the control treatment. Conversely, the E. C. variable increased by 61% across the treatments with urea compared to the control (Figure 1). In agricultural soils, measuring soil pH and E. C. is crucial, as these variables can predict nutrient availability for plants in the soil. Generally, E. C. shows a negative correlation with pH since soil salinity tends to be lower when the soil is more acidic, which can be controlled with fertilizers (Ramírez *et al*., 2022). However, some fertilizers can contribute to salinity issues when concentrated in the root zone of crops (Mata-Fernandez *et al*., 2014). Additionally, it has been reported that using urea as a nitrogen source for the remediation of soils contaminated by hydrocarbons at a dosage of 1.2 kg restored E. C. and reduced pH to 7.75, indicating that urea acts as a nitrogen source for microorganisms that decompose the contaminants present in the soils (Parillo, 2024).

Moreover, the nitrogen contained in urea reacted quickly and was efficiently expressed in the plant. Comparing the nitrogen content in the soil with that in the leaves, the concentration increased by 18% on average across the three treatments supplied with urea, which doubled the concentration compared to the control for both soil and leaves (Figure 2). This was attributed to the inherent nature of urea fertilizer, the high mobility of nitrogen within the plant, and possibly environmental conditions such as temperature, alkaline pH, and specific catalysts, especially the enzyme urease. Potassium was also influenced by urea's effect; although potassium was not supplied, the observed increase corresponded to a 90% increase in both soil and leaf concentrations on average across the three treatments supplied with urea compared to the control, where potassium concentration in soil doubled and in leaves surpassed the control by approximately 50% (Figure 2). It is probable that the increase in potassium concentration, even without direct application, was sourced from existing soil reserves, and the change in pH facilitated its absorption by the plant. It is important to emphasize that potassium is one of the most abundant cations in plant cells and is absorbed in large quantities. Once it enters the plant root, it is transported to various organs, being highly mobile. Despite the values obtained reflecting ranges between 1396 ppm (0.13%) to 4416 ppm (0.44%), for nitrogen and 245 ppm (0.024%) to 5856 ppm (0.58%) for potassium across the four treatments, these values were relatively low compared to nutrient analysis results from a study on Mexican lime in the Apatzingan Valley, where concentration determinations were as follows: for nitrogen (%): < 1.81 (deficient = 18100 ppm); 1.82-2.49 (low = 18200-24900 ppm); 2.50-3.17 (optimal = 25000-31700 ppm); 3.18-3.85 (high = 31800-38500 ppm); >3.86 (excess = 38600 ppm). For potassium (%): < 0.06 (deficient = 600 ppm); 0.07-1.27 (low = 700-12700); 1.28-1.87 (optimal = 12800-18700 ppm); 1.88-2.47 (high = 18800-24700 ppm); >2.48 (excess = 24800 ppm) (Maldonado *et al.*, 2001). This opens the possibility of exploring higher nutrient dosing strategies. Additionally, nutrient requirement determinations can be supported through the balance index analysis, a technique developed by Kenworthy (1961), which involves using the average concentration of sampled leaves from trees exhibiting a desirable horticultural development to generate a standard. This standard is established by selecting 10% or more of a population showing a desired attribute (yield) with a coefficient of variation (C. V.) below 34%. The established standard for Mexican lime in the Apatzingan Valley was: nitrogen 2.8% (C.V. 18.9) and potassium 1.57% (C.V. 23.2).

Regarding development variables, a 32% increase in cumulative bud length was observed across the average of treatments supplied with urea compared to the control; for the number of buds, an increase of 14% was noted for treatments supplied with urea over the control; and for yield per plant, the control was surpassed by 65% by the average yield from treatments supplied with urea. This confirms the positive response of Mexican lime to nitrogen supply, particularly with the characteristics of urea. This is similar to findings in a study on the interaction of factors affecting nitrate/ammonium/urea ratios and potassium concentration in nutrient solutions for tomatoes, where significant effects on development variables were found (Parra-Terraza *et al.*, 2010). In hydroponically grown tomatoes, it was also found that substituting 15% of total nitrate nitrogen in the nutrient solution with a similar percentage of ammonium and urea nitrogen or a mixture of 7.5% ammonium nitrogen and 7.5% urea nitrogen could be alternated without affecting yield (Parra *et al.*, 2012).

Finally, the yield obtained per plant for treatment 1 was 78.5 kg, for treatment 2 was 94.25 kg, for treatment 3 was 112.5 kg, and for the control was 57.5 kg. Although this yield is still below the potential yield of the species (Maldonado *et al.*, 2001), improvements can be made based on the species' response to urea fertilizer. However, it is also important to highlight the effect on pH and E. C.

variables, as they contribute significantly to soil chemical correction through fertilization, which can influence other agronomically relevant physicochemical variables.

CONCLUSIONS

The soil characteristics reflected in the chemical variables (pH, E. C., N, and K) were significant and positive for the production of lime, and the treatments supplemented with nitrogen clearly outperformed the control. Additionally, the development variables, including bud length and number of buds, were influenced by the application of urea. The concentration of nitrogen in the leaves, as well as potassium, exhibited a progressive behavior statistically influenced by the constant physiological activity of the tree. Notably, the response of potassium, despite not being supplied, reflected increases in concentration from the soil to the leaves. Among the evaluated treatments, the best yields were obtained in the treatments supplemented with nitrogen, with treatment 3 corresponding to the dose of 125 g of urea per plant, yielding 112.5 kg per plant.

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