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Effect of foliar spray of plant growth regulators and micronutrients on physio-chemical analysis of acid lime (*Citrus aurantifolia* Swingle) cv. Ganganagar lime-1

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Abstract
To determine the impact of foliar spraying plant growth regulators and micronutrients on the physio-chemical analysis of acid lime (*Citrus aurantifolia* Swingle) cv. Ganganagar Lime-1, the investigation was conducted in 2020–2021 at the experimental orchard, Department of Fruit Science, Banda University of Agriculture and Technology, Banda. The experiment was set up in RBD (Randomize Block Design) having 10 treatments with 3 replications which were treated with different concentrations of growth regulators and micronutrients. Among the plant growth attributes, the maximum initial fruit set (75.04%) and final fruit set (53.66%) were observed with the treatment T_{10} - GA_3 (75 ppm) + NAA (100 ppm) + $ZnSO_4$ (1%) + $FeSO_4$ (1%). Whereas the maximum number of seeds / fruit (6.10) were recorded similar with the treatment T_4 - (GA_3 75 PPM + $ZnSO_4$ 1%) and T_{10} - GA_3 (75 ppm) + NAA (100 ppm) + $ZnSO_4$ (1%) + $FeSO_4$ (1%) and the highest Rind thickness (mm) (1.98) reported with the treatment T_2 - GA_3 (75 ppm) + NAA (100 ppm). The other fruit quality attributes like maximum total sugars (1.66%) and reducing sugars (0.88%) were also recorded with the treatment T_{10} - GA_3 (75 ppm) + NAA (100 ppm) + $ZnSO_4$ (1%) + $FeSO_4$ (1%). The spray of GA_3 (75 ppm) + NAA (100 ppm) + $ZnSO_4$ (1%) + $FeSO_4$ (1%) can be advised to farmers and researchers to achieve maximum fruit yield and good fruit quality in Ganganagar Lime-1 cultivar of acid lime based on the results of foliar spray of plant growth regulators and micronutrients on growth, fruit yield, and quality of acid lime under Bundelkhand region of Uttar Pradesh.

Keywords: Fruit quality, growth regulators, micronutrients, orchard and randomize block design

1. Introduction
More than 162 species with the chromosomal number (2n=18) and belonging to the family Rutaceae and order Gerenials are included in the genus Citrus. Several fruits, including acid lime, sweet lime, mandarin, grape fruit, pummelo, etc., are referred to as "citrus" in this context. It is a significant fruit in India and among the most cultivated worldwide. The majority of citrus species are indigenous to South East Asia's tropical and subtropical climates, mainly India, China, and the area between these two nations. Few Citrus species are thought to have a native habitat in the north-eastern part of India. Next to mandarin and sweet orange, acid lime (*Citrus aurantifolia* Swingle) is the third most significant fruit crop. It has spread around the world since its inception in India and is widely known as "lime" or "Nimbu" in the native tongue. Limes require a tropical, dry climate with an ideal range of 20 to 30 °C. Low humidity encourages better colour development, whereas high humidity makes fruit more juicy with thin rinds (Cooper *et al*., 1963) [2]. Humidity plays a significant effect in fruit growth. Almost all states, including Andhra Pradesh, Bihar, Assam, Jharkhand, and Chhattisgarh, grow it. The lime fruit has a straightforward sigmoid development pattern and is categorised as a non-climacteric fruit since it releases very little ethylene as it ripens and has significant medicinal potential. Acid lime blooms all year long in the three separate "Bahar" seasons of Ambe, Mrig, and Hasth. The size of fruits remains often quite tiny and fruit quality characteristics are further impacted by continual flowering and excessive crop loads on trees, leading to the harvest of subpar and unmarketable fruits. Plant growth regulators can be used successfully to promote fruit set since insufficient pollination also affects fruit set. Micronutrients and plant growth regulators are very important for many physiological processes in plants, and foliar administration of these substances is more effective than soil

application. They are used to control blooming, thin down flowers and fruits, increase fruit set and size, decrease pre-harvest fruit drop, and promote fruit growth. They serve as a metabolic sink for the transfer of metabolites between different parts of the plant, particularly towards growing fruits. Plant growth regulators used close to a tree's terminal buds may accelerate development by encouraging more or less continuous growth throughout the growing season. In addition to stimulating cell division and elongation, GA_3 application also affects citrus blooming. The production of additional carbohydrates in the leaves is increased by iron, which also controls fruit drop and promotes flowering, fruit set, fruit size, and plant output. It may be found in ferrodoxin, which precipitates in oxidation-reduction reactions like (NO4-) and (SO4-) reduction, N-fixation, and others, as well as in numerous flavo-proteins, peroxidase, catalase, and cytochrome oxidase enzymes. More than 150 enzymes and hormones, including growth hormones and enzymes involved in respiration, are composed of or activated by zinc. In addition, zinc has a role in the synthesis of enzymes that produce auxins in plants. Numerous enzymes with specificity in the synthesis of carbon dioxide are also activated by zinc. In some enzymes, such DNA (deoxyribonucleic acid) polymerase and alcohol dehydrogenase, zinc (Zn) has a beneficial function. It boosts the chlorophyll content of leaves and is crucial for the enzymatic processes required for fruit growth and development. According to Sharma *et al.* (2008), Zn is also involved in controlling the metabolism of proteins and carbohydrates. Thus encourages the synthesis of IAA through tryptophan, which acts as a precursor for the synthesis of auxins, and thus directly influences the growth parameters. The lime acid cv. Ganganagar Lime - 1 is an early and prolific bearer with a rich yellow fruit colour, fragrance, high juice content, few seeds (5–6), a thin skin, canker resistance, and great yield. In the Bundelkhand area of Uttar Pradesh, Ganganagar Lime - 1 has smaller fruits than other commercial lime types because of its high output. Fruit size also affects how marketable they are. The present experiment was carried out to investigate the impact of foliar application of plant growth regulators and micronutrients on growth, fruit yield, and quality of acid lime cv. It was done so in light of the need to increase the fruit size of this cultivar for this region and the significant role of plant growth regulators and micronutrients as mentioned above. Lime from Ganganagar: 1.

2. Materials and Methods

The current study, titled "Effect of Foliar Spray of Plant Growth Regulators and Micronutrients on Physio-Chemical Analysis of Acid Lime (*Citrus aurantifolia* Swingle) cv. Ganganagar Lime-1," was conducted in the years 2020–2021 at the experimental orchard on the campus of the Department of Fruit Science, Banda University of Agriculture and Technology, Banda. The following is a description of the specific materials utilised, chosen techniques, recorded observations, and statistical analysis used during the current investigation:

2.1 Analysis of data

The experiment's two years' worth of data were statistically examined using Panse and Sukhatme's (1985) [7] methodology, and findings were assessed at a 5% level of significance. The following formula was used to calculate the standard error (S.Em.±) for the difference in treatment means.

Where;
MSE = Mean sum of squares due to error r = Number of replications.
The calculation of C.D. at 5% of table value was carried out with the help of following formula.

$$\mathbf{C.D.} = \mathbf{S.Em.} \pm \sqrt{\mathbf{2}} \times \mathbf{t\ value\ at\ 5\%}$$

C.D. = Critical difference
S.Em.± = Standard error of mean.

2.2 Treatment details

The experimental trees were subjected to the treatments comprising of the combinations of plant growth regulators and micronutrients. The detail of treatment is presented in table number.

Table 1: Treatment details of the experiment.

Treatment codes	Detail of the treatments
T_1	Control (water spray)
T_2	GA_3 (75 ppm) + NAA (100 ppm)
T_3	$ZnSO_4$ (1%) + $FeSO_4$ (1%)
T_4	GA_3 (75 ppm) + $ZnSO_4$ (1%)
T_5	GA_3 (75 ppm) + $FeSO_4$ (1%)
T_6	NAA (100 ppm) + $ZnSO_4$ (1%)
T_7	NAA (100 ppm) + $FeSO_4$ (1%)
T_8	GA_3 (75 ppm) + $ZnSO_4$ (1%) + $FeSO_4$ (1%)
T_9	NAA (100 ppm) + $ZnSO_4$ (1%) + $FeSO_4$ (1%)
T_{10}	GA_3 (75 ppm) + NAA (100 ppm) + $ZnSO_4$ (1%) + $FeSO_4$ (1%)

[Where, GA_3 = Gibberellic acid, NAA = Naphthalene acetic acid, $ZnSO_4$ = Zinc sulphate, $FeSO_4$ = Iron sulphate.]

The data on the growth, yield and fruit quality parameters were recorded and the procedures followed are described below:

2.2.1. Initial fruit set (%)

Number of flowers were counted and tagged. After a month number of setted fruits were counted and divided by total number of flowers. The initial fruit set was expressed in percent with help of following formula:

$$\text{Initial fruit set (\%)} = \frac{\text{Number of setted fruit}}{\text{Total number of flower}} \times 100$$

2.2.2. Final fruit set (%)

The number of fruit sets was counted, their total was divided by the total number of fruits harvested, and the result was stated as a percentage using the following formula:

$$\text{Final fruit set (\%)} = \frac{\text{Number of fruits at the time harvest}}{\text{Number of setted fruits}} \times 100$$

2.2.3. Number of seeds/fruit

From 10 smashed fruits, seeds were taken. The seeds were then properly cleaned with water after being gathered in a beaker. Then, 10 fruits were averaged to determine the average amount of seeds per fruit.

2.2.4. Rind thickness

Fruits were sliced into equal halves using a sharp knife to measure the thickness of the rind, which was then measured in millimetres with the use of a digital vernier calliper.

2.2.5. Total sugars

20 ml of acid lime juice and 5 ml of concentrated HCL (hydrochloric acid) were taken in a beaker for the estimate of total sugars. The solution was then heated over a water bath for five minutes to cause hydrolysis, which turns the non-reducing sugars into reducing sugars. After cooling, sodium carbonate solution was used to neutralise any surplus acid. The solution was put into a 100 ml volumetric flask, and the required amount of distilled water was added to make the volume correct. The Fehling's solutions "A" and "B" were used to titrate the solution after it was taken in a burette. similar to how sugars were reduced (Nelson, 1994). The following calculation was used to compute the overall sugar percentage:

$$\text{Total sugars (\%)} = \frac{0.25}{\text{Burette reading}} \times 100$$

2.2.6. Reducing sugars

The approach outlined by Nelson (1994) was used to estimate the reduction of sugars in fruit juice. In a 300 ml conical flask, 5 ml of Fehling's "A" and "B" solutions from each were collected and diluted with 40 ml of distilled water. The juice solution was added gradually to the heated, boiling Fehling's solution until a faint crimson hue appeared. Three drops of methylene blue indicator were now added, and the titration process was maintained until a brick red precipitate formed and completely obliterated the blue colour. The following equation was used to compute the proportion of reducing sugars:

$$\text{Reducing sugars (\%)} = \frac{+0.25}{\text{Burette reading}} \times 100$$

3. Results

3.1 Initial fruit set (%)

The results in Table 2 showed that the treatment T_{10} (GA_3(75 PPM) + NAA (100 PPM) + $ZnSO_4$ (1%) + $FeSO_4$ (1%) had the highest initial fruit set (75.04%), which was statistically comparable to the treatments T_9 (NAA (100 PPM) + $ZnSO_4$ (1%) + $FeSO_4$ (1%), T_2 (GA_3(75 PPM + NAA (100 PPM), T_6 (NAA 100 PPM + Z The treatment T_1 (Control) was found to have the lowest percentage of fruit set (61.35%).

3.2 Final fruit set (%)

It is clear from the data presented in table 2 that the maximum final fruit set (53.66%) was recorded with the treatment T_{10} (GA_3 (75 PPM) + NAA (100 PPM) + $ZnSO_4$ (1%) + $FeSO_4$ (1%) which was statistically at par with the treatment T_9 (NAA (100 PPM) + $ZnSO_4$ (1%) +$FeSO_4$ (1%) and was followed by the T_2 (GA_3 (75 PPM) +NAA (100 PPM) that recorded final fruit set percentage of 51.61% and 49.70% respectively. Additionally, the treatment T_1 (Control) produced the highest ultimate fruit set (32.27%).

Table 2: Acid lime (*Citrus aurantifolia* Swingle) cv. Ganganagar Lime - 1 initial fruit set and final fruit set in response to plant growth regulators and micronutrients

Treatments	Initial fruit set (%)	Final fruit set (%)
T_1 (Control)	61.35	32.27
T_2 (GA_3 75 PPM + NAA 100 PPM)	73.55	49.70
T_3 ($ZnSO_4$ 1% + $FeSO_4$%)	64.61	34.58
T_4 (GA_3 75 PPM + $ZnSO_4$ 1%)	67.60	38.54
T_5 (GA_3 75 PPM + $FeSO_4$ 1%)	66.83	36.63
T_6 (NAA 100 PPM + $ZnSO_4$ 1%)	72.51	44.45
T_7 (NAA 100 PPM + $FeSO_4$ 1%)	70.86	41.32
T_8 (GA_3 75 PPM + $ZnSO_4$ 1% + $FeSO_4$ 1%)	68.55	39.46
T_9 (NAA 100 PPM + $ZnSO_4$ 1% + $FeSO_4$ 1%)	74.26	51.61
T_{10} (GA_3 75 PPM + NAA 100 PPM + $ZnSO_4$ 1% + $FeSO_4$ 1%))	75.04	53.66
C.D.at 5%	2.65	2.97
S. Em. ±	0.88	0.99

3.3 Number of seeds per fruit

The data presented in Table 3 that shows that there was non-significant effect of plant growth regulators and micronutrients on the number of seeds per fruit of acid lime cv. Ganganagar Lime – 1.

3.4 Rind thickness (mm)

The results showed that the treatment of plants with growth regulators and micronutrients had a substantial impact on the rind thickness of acid lime (*Citrus aurantifolia* Swingle) cv. Ganganagar Lime - 1. The results are shown in Table 3. The treatment T_2 (GA_3 (75 PPM) + NAA (100 PPM)) produced the thickest rind (1.98 mm). With the treatment T_8 (GA_3 (75 PPM) + $ZnSO_4$ (1%) + $FeSO_4$ (1%), the rind thickness (1.66 mm) was measured. The results also showed that treatment T_8, which recorded a rind thickness of 1.70 mm and mm, respectively, was statistically comparable to treatment T_9 (NAA (100 PPM) + $ZnSO_4$ (1%) + $FeSO_4$ (1%).

Table 3: Effect of plant growth regulators and micronutrients on the number of seeds per fruit and rind thickness of acid lime (*Citrus aurantifolia* Swingle) cv. Ganganagar Lime – 1

Treatments	Number of seeds / fruit	Rind thickness (mm)
T_1 (Control)	6.03	1.76
T_2 (GA_3 75 PPM + NAA 100 PPM)	5.96	1.98
T_3 ($ZnSO_4$ 1% + $FeSO_4$%)	6.03	1.91
T_4 (GA_3 75 PPM + $ZnSO_4$ 1%)	6.10	1.83
T_5 (GA_3 75 PPM + $FeSO_4$ 1%)	6.06	1.87
T_6 (NAA 100 PPM + $ZnSO_4$ 1%)	6.03	1.80
T_7 (NAA 100 PPM + $FeSO_4$ 1%)	6.10	1.85
T_8 (GA_3 75 PPM + $ZnSO_4$ 1% + $FeSO_4$ 1%)	5.83	1.66
T_9 (NAA 100 PPM + $ZnSO_4$ 1% + $FeSO_4$ 1%)	5.90	1.70
T_{10} (GA_3 75 PPM + NAA 100 PPM + $ZnSO_4$ 1% + $FeSO_4$ 1%))	6.03	1.73
C.D.at 5%	NS	0.08
S. Em.±	0.06	0.02

3.5 Total sugar (%)
The maximum percentage of total sugar (1.66%) plus $FeSO_4$ (1%), which was statistically comparable to the treatments T_9 (NAA (100 PPM) + $ZnSO_4$ (1%), T_8 (GA_3 (75 PPM) + $ZnSO_4$ (1%), and T_2 (GA_3 (75 PPM) + NAA (100 PPM), which recorded total sugars of 1.60%, 1.53%, and 1.50% respectively, was shown in Table 4. The treatment T_1 (control) was found to have the lowest amount of total sugars (1.06%).

3.6 Reducing sugars (%)
Based on a review of the information in Table 4. The maximum percentage of reducing sugar (0.88%) was recorded with treatment T_{10} (GA_3 (75 PPM) + NAA (100 PPM) + $ZnSO_4$ (1%) + $FeSO_4$ (1%) which was statistically at par with the treatment T_9 (NAA (100 PPM) + $ZnSO_4$ (1%) + $FeSO_4$ (1%) and was followed by treatment T_8 (GA_3 (75 PPM) + $ZnSO_4$ (1%) + $FeSO_4$ (1%) that recorded reducing sugar percentage of 0.87% and 0.85% respectively. The treatment T_1 (Control) was found to have the lowest level of reducing sugars (0.70%).

Table 4: Effect of plant growth regulators and micronutrients on the total sugar and reducing sugar of acid lime (*Citrus aurantifolia* Swingle) cv. Ganganagar Lime – 1

Treatments	Total sugar (%)	Reducing sugar (%)
T_1 (Control)	1.06	0.70
T_2 (GA_3 75 PPM + NAA 100 PPM)	1.50	0.84
T_3 ($ZnSO_4$ 1% + $FeSO_4$%)	1.16	0.74
T_4 (GA_3 75 PPM + $ZnSO_4$ 1%)	1.40	0.82
T_5 (GA_3 75 PPM + $FeSO_4$ 1%)	1.20	0.76
T_6 (NAA 100 PPM + $ZnSO_4$ 1%)	1.43	0.83
T_7 (NAA 100 PPM + $FeSO_4$ 1%)	1.33	0.78
T_8 (GA_3 75 PPM + $ZnSO_4$ 1% + $FeSO_4$ 1%)	1.53	0.85
T_9 (NAA 100 PPM + $ZnSO_4$ 1% + $FeSO_4$ 1%)	1.60	0.87
T_{10} (GA_3 75 PPM + NAA 100 PPM + $ZnSO_4$ 1% + $FeSO_4$ 1%))	1.66	0.88
C.D.at 5%	0.22	0.02
S. Em.±	0.07	0.009

4. Discussion
The current studies on "Effect of foliar spray of plant growth regulators and micronutrients on physio-chemical analysis of acid lime (*Citrus aurantifolia* Swingle) cv. Ganganagar Lime - 1" were carried out to assess the impact of plant growth regulators and micronutrients on acid lime.

It is clear from the current study that micronutrients and plant growth regulators have an impact on fruit set and production (Table 2). When T_{10} [GA_3 (75 ppm) + NAA (100 ppm) + $ZnSO_4$ (1%) + $FeSO_4$ (1%)] was applied, the highest percentages of initial fruit set (75.04%) and ultimate fruit set (53.66%) were observed. The application of GA_3, which is thought to serve as a mediating process for faster translocation and metabolization of stored metabolites and photosynthates from source to sink and also play a significant role in increasing the auxin synthesis in ovaries, may be the cause of the higher fruit set with the use of plant growth regulators (Jagtap *et al*., 2012) [3]. Pollen germination and pollen tube development may have been impacted by the greater fruit set brought on by the use of GA_3. Similar to this, treating micronutrients with zinc sulphate and iron sulphate boosted acid lime fruit set and yield. Higher levels of zinc sulphate raised the zinc content of the foliar tissue, which in turn promoted the endogenous synthesis of auxin, which decreased fruit loss and promoted fruit retention (Krishnamoorthy, 1992) [5]. As tryptophan is a precursor to endogenous auxin and auxin prevents abscission and makes it easier for the ovary to stay attached to the shoots, zinc is crucial for preventing the formation of the abscission layer (Jat and Kacha, 2014) [4]. This reduces flower and fruit drop. Yadav *et al*. (2011) [8] and Bhambota *et al*. (1962) [1] both came to similar conclusions. Treatment with growth regulators and micronutrients had a substantial impact on fruit quality (Table 4). The results showed that the treatment T_{10} [GA_3 (75 ppm) + NAA (100 ppm) + $ZnSO_4$ (1%) + $FeSO_4$ (1%)] produced the highest levels of total sugars (1.66%) and reducing sugars (0.88%).

5. Conclusion
According to the findings of the current experiments, treatment T_{10} [GA_3 (75 ppm) + NAA (100 ppm) + $ZnSO_4$ (1%) + $FeSO_4$ (1%)] displayed the best fruit physico-chemical characteristics among the various concentrations of plant growth regulators and micronutrients. Therefore, the spray of GA_3 (75 ppm) + NAA (100 ppm) + $ZnSO_4$ (1%) + $FeSO_4$ (1%) can be advised to farmers and researchers to obtain maximum fruit yield and good fruit quality in Ganganagar Lime -1 cultivar of acid lime based on results of foliar spray of plant growth regulators and micronutrients on fruit growth and fruit quality of acid lime under Bundelkhand region of Uttar Pradesh.

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