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## EFFECT OF BIO-FERMENTED ORGANIC FERTILIZER AND FOLIAR APPLICATION OF SOME PLANT EXTRACTS ON THE GRAPE (*VITIS VINIFERA* L.)

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### SUMMARY

The following study aimed to examine the effects of EM Bokashi leaves (a biofertilizer) and several plant extracts in three distinct grape (*Vitis vinifera* L.) cultivars. The experiment comprised three components: the first was the three grape cultivars (Summer Royal, Flame, and Crimson); the second was the two different doses of EM Bokashi (250 g per seedling); and the third was the application of plant extracts at four distinct concentrations—no spraying and 15 ml L<sup>-1</sup> each of the licorice root powder, roselle petals, and moringa leaves. The experiment adopted a factorial design using a randomized complete block design (RCBD). The seedless cultivar Summer Royal resulted in a triple interaction with EM Bokashi at a dosage of 250 g. The interaction of seedling<sup>-1</sup> with the foliage of aqueous moringa leaf extracts at a concentration of 15 ml L<sup>-1</sup> increased the volume of both components in the leaves. The iron and boron concentration, together with the total carbohydrate content in leaves' auxins and cytokines, was comparable to the Crimson cultivar, which did not include effective microorganisms (EM Bokashi). However, this treatment exhibited the lowest average values for the examined features.

**Keywords:** Grape (*V. vinifera* L.), cultivars, bio-fermented organic fertilizer, plant extract, biochemical composition

**Key findings:** The EM Bokashi treatment at 250 g boosted the quality of the evaluated grapes (*V. vinifera* L.), while also increasing the amount of spraying with moringa leaf extract at 15 ml per liter. The findings indicated the potential of seedless grape varieties to adapt to diverse experimental circumstances.

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## INTRODUCTION

The grape (*Vitis vinifera* L.) is a member of the genus *Vitis*, which is part of the family Vitaceae, comprising 14 genera (Dhafer, 2022). Grapes are native to every part of the world, and they usually adapt to different environments, particularly in temperate climates that are tropical or have other similarities. The estimated number of grapevines in Iraq is 11,613,783, and they produce an annual volume of 421,868 tons, with a typical yield of 36.32 kilograms per plant (CSO, 2020).

The use of microbial and organic fertilizers has become popular in agriculture globally to help reduce the negative effects on plants, people, and the environment. This includes the use of organic materials, such as biofertilizers and fertilizers. Among these fertilizers is Bokashi, an organic fertilizer with potential for chemical processing. Numerous microbes have been shown to colonize the root zone and aid in plant growth by boosting nutrient availability, restoring organic matter, fortifying soil properties, and enhancing moisture retention (Karimuna *et al.*, 2016).

Foliar fertilization tended to be the most important component of the strategy because it leads to the greatest degree of nutrient absorption compared with soil fertilization. Studies have demonstrated that plants can receive as much as 85% of the nutrients they need via foliar coating (Al-Sahhaf, 1989). Among these nutrients, non-commercial plant extracts are often applicable as biostimulants or pesticides, containing massive bioactive compounds, primarily phenols, phytohormones, amino acids, and vitamins. Additionally, they have a high concentration of macro- and micronutrients. The composition of these active ingredients is dependent on the plant species, growth conditions, the parts used from the plant for extraction, and the method of extraction. Examples of extracts that contain these ingredients include root licorice, gooseberry, moringa, onion, garlic, turmeric, and ginger (Bulgari *et al.*, 2019).

Despite the beneficial environmental conditions in Iraq and the lack of diverse

methods of grape cultivation, their cultivation is still modest. These cultivars are appropriate for consumption as table grapes, dried grapes (raisins), or juices. Additionally, they are seedless with high sugar content, which entices consumers (Akkurt *et al.*, 2019). As such, it is essential to focus on these seedless cultivars and investigate ways to increase their production in Iraq via organic methods using fertilizers and the application of specific extracts via foliage. American cultivars of seedless grapes include Summer Royal, Flame, and Crimson. This investigation intended to explore the effects of the EM-Bokashi fertilizer and the foliage of plant extracts regarding three different grape cultivars, with primary focus on their chemical properties.

## MATERIALS AND METHODS

The study on grapes (*V. vinifera* L.) took place in a slatted barn on a private farm near Al-Musaib District between September 2022 and July 2023. The research examined the effects of foliar-sprayed EM Bokashi along with various plant extracts on three different American grape cultivars. The three seedless grape cultivars used in the first component of the experimental design consisted of Summer Royal, Flame, and Crimson as the types undergoing analysis. Two increases in EM Bokashi comprised the second component: one with no addition and one with 250 ml added per seedling. Aqueous extracts at 15 ml/L each of licorice roots, roselle petals, and moringa leaves and no spraying were the four concentrations of plant extracts sprayed on the leaves in the third component. With three distinct investigations ( $3 \times 2 \times 4$ ), the investigation used a factorial design, where each used a random complete block design (RCBD). Out of the 96 seedlings in each replication, four seedlings entailed assigning to each experimental group. Using an automatic sprinkler system helped provide a steady coating of water on the vegetative seedlings, with the spraying done early in the morning. Only distilled water was the water type given to the seedlings in the control group, which had 288 seedlings.

## **Studied characteristics**

### **Leaf iron and boron content**

The mineral composition determination transpired in the laboratory of the Department of Plant Production Engineering, Al-Musaib Technical College. The randomly selected adult leaves from each experimental group incurred soaking in water with a high degree of purity. Then, placing the samples in paper bags with holes sustained oven drying at 70 °C for 48 h until the weight became steady. After drying the samples, they underwent grinding using an electric mill. Then, taking 0.5 g of each sample succeeded in digesting by adding sulfuric acid and perchloric acid to a colorless extract that aided in determining the minerals (Nada, 2022).

### **Leaf total carbohydrates**

The determination of the total amount of carbohydrates proceeded by extracting soluble sugars according to the method of Joslyn (1970). The 200 mg of dried and previously ground cotyledons from each treatment, upon adding to a test tube, received 8 ml of 80% ethanol. Then, the tube, placed in a 60 °C water bath for 30 min with constant stirring, underwent centrifuging to determine the percentage of reducing sugars and starch in the sample using a spectrophotometer at a 480 nm wavelength.

### **Leaf auxin and cytokinin contents**

Auxin quantification used a one-gram sample of leaves to which adding 12 ml of methanol, 5 ml of chloroform, and 3 ml of hydroxide ensued for dry weight. The volume alteration to 25 ml comprised adding distilled water there, with the acidity of the solution set to pH 2.5 by adding hydrochloric acid (1 liter) in a single dose. The study of samples employed a spectrophotometer with a wavelength of 222 nm for auxin. A standard graph construction continued for each hormone and recorded the results (Nuray *et al.*, 2002).

For cytokinins, collecting the lower portion of the mixture (12 ml of methanol, 5

ml of chloroform, and 3 ml of ammonium hydroxide) succeeded, altering the pH to 7, and measuring cytokinins at a wavelength of 269 nm (Nuray *et al.*, 2002).

## **Statistical analysis**

The data analysis statistically used ANOVA, based on RCBD with a rectangular configuration. The most significant difference ( $LSD_{0.05}$ ) as employed contrasted and isolated means (Gomez and Gomez, 1984) engaging the statistical programming software GenStat12.

## **RESULTS**

### **Leaf iron content**

The grape (*V. vinifera* L.) cultivars indicated significant differences for iron content in the leaves (Table 1). The Summer Royal cultivar had a premier increase in iron content, with a value of 194.98 mg kg<sup>-1</sup>, while the Crimson cultivar had the lowest at 120.88 mg kg<sup>-1</sup>. The same chart also demonstrates the application of 250 g of EM Bokashi to the ground remarkably enriched the iron content of the soil. The maximum value of the treatment was 182.52 mg kg<sup>-1</sup>, while the minimum value of the control treatment was 152.28 mg kg<sup>-1</sup>. The leaves sprayed with extracts from plants experienced a significant rise in the iron content: the content of iron in moringa leaves was 175.14 mg kg<sup>-1</sup>, while the minimum value of the control treatment was 152.73 mg kg<sup>-1</sup>.

Results from the same table exhibited a significant correlation between the kind of EM Bokashi and soil utilization. Although the least effective treatment was just 116.42 mg/kg, the 250 g therapy was very successful at 230.63 mg/kg. The cultivars Summer Royal and moringa extracts had the highest concentration of iron in their leaves (208.90 mg/kg), while the unsprayed Crimson cultivar had the lowest concentration (112.25 mg/kg), according to the interaction between the cultivar and the foliar application of plant extracts. The 250 g of EM Bokashi plus the moringa leaf extract treatment had an iron level of 189.43 mg/kg,

**Table 1.** Effect of Bokashi EM and foliar spraying with some plant extracts for three grape cultivars and their interactions on the leaf iron content ( $\text{mg kg}^{-1}$ ).

| Cultivars                                         | EM Bokashi   | Foliar spraying with plant extracts |                       |                              |                               | Interaction between cultivars and Bokashi |
|---------------------------------------------------|--------------|-------------------------------------|-----------------------|------------------------------|-------------------------------|-------------------------------------------|
|                                                   |              | Control                             | Licorice root         | Moringa                      | Roselle petals/leaves         |                                           |
| Summer Royal seedless                             | control      | 121.30                              | 170.84                | 172.20                       | 172.93                        | 159.32                                    |
|                                                   | EM Bokashi   | 204.97                              | 240.43                | 245.60                       | 231.53                        | 230.63                                    |
| Flame seedless                                    | control      | 177.84                              | 181.92                | 184.22                       | 180.48                        | 181.11                                    |
|                                                   | EM Bokashi   | 187.80                              | 191.63                | 195.67                       | 191.30                        | 191.60                                    |
| Crimson seedless                                  | control      | 100.21                              | 120.12                | 126.15                       | 119.21                        | 116.42                                    |
|                                                   | EM Bokashi   | 124.30                              | 125.12                | 127.03                       | 124.96                        | 125.33                                    |
|                                                   |              |                                     |                       |                              |                               | Effect of cultivars                       |
| Interaction between cultivars and plant extracts  | Summer Royal | 163.13                              | 205.64                | 208.90                       | 202.23                        | 194.98                                    |
|                                                   | Flame        | 182.82                              | 186.77                | 189.94                       | 185.89                        | 186.36                                    |
|                                                   | Crimson      | 112.25                              | 122.59                | 126.59                       | 122.08                        | 120.88                                    |
|                                                   |              |                                     |                       |                              |                               | Effect of EM Bokashi                      |
| Interaction between plant extracts and EM Bokashi | control      | 133.11                              | 157.63                | 160.86                       | 157.54                        | 152.28                                    |
|                                                   | EM Bokashi   | 172.36                              | 185.71                | 189.43                       | 182.60                        | 182.52                                    |
| effect of spraying with plant extracts            |              |                                     |                       |                              |                               |                                           |
| LSD 0.05                                          |              |                                     |                       |                              |                               |                                           |
| cultivars                                         | EM Bokashi   | plant extracts                      | cultivars and Bokashi | cultivars and plant extracts | plant extracts and EM Bokashi | triple interaction                        |
|                                                   |              |                                     |                       |                              |                               |                                           |
| 1.08                                              | 0.88         | 1.24                                | 1.52                  | 2.15                         | 1.76                          | 3.04                                      |

whereas the control treatment had a lower iron level of  $133.11 \text{ mg/kg}$ . This interaction between EM Bokashi and plant extracts led to a substantial increase in iron levels. The iron levels in leaves received considerable impacts from the triple treatment combination. With a weight of 250 g, the Summer Royal cultivar treatment produced the topmost value at  $245.60 \text{ mg kg}^{-1}$ , whereas the control of EM and plant extract treatments produced the lowest value at  $100.21 \text{ mg kg}^{-1}$ .

### Leaf boron content

Grape cultivars showed varied values for boron content in the leaves (Table 2). At  $29.60 \text{ mg}$  of boron per kg, the Summer Royal cultivar had a much higher boron content than the Crimson cultivar, while the latter had the lowest concentration at  $16.62 \text{ mg per kg}$ . Following the application of EM Bokashi 250 g to the plant, the amount of boron increased significantly. In the control treatment, the lowest result was  $21.28 \text{ mg kg}^{-1}$ , whereas the highest value generated by EM Bokashi was

$25.13 \text{ mg kg}^{-1}$ . Plant extracts applied topically boosted the plants' boron content: the leaf extract from moringa had the maximum concentration at  $24.38 \text{ mg kg}^{-1}$ , while the control had the minimum at  $21.41 \text{ mg kg}^{-1}$ . The results detailed in the same table indicate the type of soil and the EM Bokashi fertilizer significantly impacted how treatments interacted. The Crimson cultivar treatment without additives had the lowest effect at  $15.88 \text{ mg/kg}^{-1}$ , while the Summer Royal and EM Bokashi therapy (250 g) had the most influence at  $33.98 \text{ mg/kg}^{-1}$ . The treatment with Summer Royal and moringa leaf extract produced the highest concentration of boron in leaves at  $32.03 \text{ mg kg}^{-1}$ , while the treatment with Crimson cultivar without a spray produced the lowest concentration at  $16.11 \text{ mg kg}^{-1}$ . This was the result of the interaction between the cultivar and the foliar application of plant extracts. The boron content rose significantly when applying plant extracts and the interaction between EM Bokashi and the leaves. The control treatment yielded a lower result of  $20.38 \text{ mg/kg}^{-1}$ , whereas the

**Table 2.** Effect of Bokashi EM and foliar spraying with some plant extracts for three grape cultivars and their interactions on the leaf boron content (mg kg<sup>-1</sup>).

| Cultivars                                         | EM Bokashi   | Foliar spraying with plant extracts |                       |                              |                               | Interaction between cultivars and Bokashi EM |
|---------------------------------------------------|--------------|-------------------------------------|-----------------------|------------------------------|-------------------------------|----------------------------------------------|
|                                                   |              | Control                             | Licorice root         | Moringa                      | Roselle petals/leaves         |                                              |
| Summer Royal seedless                             | control      | 23.58                               | 25.29                 | 25.92                        | 26.08                         | 25.22                                        |
|                                                   | EM Bokashi   | 26.94                               | 33.75                 | 37.24                        | 37.98                         | 33.98                                        |
| Flame seedless                                    | control      | 22.15                               | 22.51                 | 22.92                        | 23.39                         | 22.74                                        |
|                                                   | EM Bokashi   | 23.61                               | 23.71                 | 24.04                        | 24.89                         | 24.06                                        |
| Crimson seedless                                  | control      | 15.42                               | 15.96                 | 16.03                        | 16.13                         | 15.88                                        |
|                                                   | EM Bokashi   | 16.79                               | 16.96                 | 17.83                        | 17.82                         | 17.35                                        |
| Effect of cultivars                               |              |                                     |                       |                              |                               |                                              |
| Interaction between cultivars and plant extracts  | Summer Royal | 22.26                               | 29.52                 | 31.58                        | 32.03                         | 29.60                                        |
|                                                   | Flame        | 22.88                               | 23.11                 | 23.48                        | 24.14                         | 23.40                                        |
|                                                   | Crimson      | 16.11                               | 16.46                 | 16.93                        | 16.98                         | 16.62                                        |
| Effect of EM Bokashi                              |              |                                     |                       |                              |                               |                                              |
| Interaction between plant extracts and EM Bokashi | control      | 20.38                               | 21.25                 | 21.62                        | 21.86                         | 21.28                                        |
|                                                   | EM Bokashi   | 22.45                               | 24.81                 | 26.37                        | 26.90                         | 25.13                                        |
| effect of spraying with plant extracts            |              | 21.41                               | 23.03                 | 24.00                        | 24.38                         |                                              |
| LSD 0.05                                          |              |                                     |                       |                              |                               |                                              |
| cultivars                                         | EM Bokashi   | plant extracts                      | cultivars and Bokashi | cultivars and plant extracts | plant extracts and EM Bokashi | triple interaction                           |
| 0.70                                              | 0.57         | 0.81                                | 0.99                  | 1.40                         | 1.14                          | 1.98                                         |

treatment including 250 g of EM Bokashi and moringa leaf extract gave a value of 26.90 mg/kg<sup>-1</sup>.

The content of leaf boron ultimately appeared to acquire relevant influences from the triple treatment combinations as well; the control treatment had the lowest value of 15.42 mg/kg, while the treatment comprising Summer Royal cultivar, 250 g of EM Bokashi, and moringa leaf extract had the highest value of 37.98 mg/kg.

### Leaf total carbohydrates

The total carbohydrate content of the leaves was variable in various grape genotypes (Table 3). The variation of Summer Royal had a larger rate of 7.84%, while the cultivar of Crimson had the lowest percentage of 3.27%. The application of EM Bokashi to the soil raised the total carbohydrate content considerably, with the highest at 6.11%, while the control had a minimum of 4.57%. The total carbohydrate content of plant extracts incurs a boost of nearly 100%. The moringa leaf extract had a concentration of 6.02%, whereas the control had a minimum of 4.72%.

According to the results presented in the same table, significant differences emerged in the types of EM Bokashi and the soil application method concerning the interaction of treatments. The therapy with the Crimson cultivar without additions had the lowest effect, at just 2.27%, whereas the treatment with Summer Royal and EM Bokashi (250 g) had a prominent impact of 9.13%. When combining cultivar and foliar application of plant extracts, the treatment with Summer Royal and moringa leaf extract had the topmost total carbohydrate content in leaves (8.96%), while the treatment with Crimson cultivar and no spraying had the lowest value (2.86%). The total amount of carbohydrates increased significantly as a result of the interaction between the EM Bokashi and the foliar application of plant extracts: the treatment containing 250 g of EM Bokashi and moringa leaf extract had a higher value of 7.08% than the control group with a value of 4.03%.

**Table 3.** Effect of Bokashi EM and foliar spraying with some plant extracts for three grape cultivars and their interactions on the percentage of total carbohydrates in the leaves.

| Cultivars                                         | EM Bokashi   | Foliar spraying with plant extracts |                       |                              |                               | Interaction between cultivars and Bokashi EM |
|---------------------------------------------------|--------------|-------------------------------------|-----------------------|------------------------------|-------------------------------|----------------------------------------------|
|                                                   |              | Control                             | Licorice root         | Moringa                      | Roselle petals/leaves         |                                              |
| Summer Royal seedless                             | control      | 6.02                                | 6.77                  | 6.77                         | 6.61                          | 6.54                                         |
|                                                   | EM Bokashi   | 8.51                                | 10.28                 | 11.14                        | 6.58                          | 9.13                                         |
| Flame seedless                                    | control      | 3.74                                | 4.74                  | 4.79                         | 4.48                          | 4.45                                         |
|                                                   | EM Bokashi   | 5.23                                | 5.84                  | 5.98                         | 4.50                          | 5.39                                         |
| Crimson seedless                                  | control      | 2.33                                | 2.91                  | 3.33                         | 2.30                          | 2.27                                         |
|                                                   | EM Bokashi   | 3.38                                | 3.96                  | 4.12                         | 3.83                          | 3.82                                         |
| Effect of cultivars                               |              |                                     |                       |                              |                               |                                              |
| Interaction between cultivars and plant extracts  | Summer Royal | 7.26                                | 8.52                  | 8.96                         | 6.60                          | 7.84                                         |
|                                                   | Flame        | 4.49                                | 5.32                  | 5.39                         | 4.49                          | 4.92                                         |
|                                                   | Crimson      | 2.86                                | 3.43                  | 3.72                         | 3.07                          | 3.27                                         |
| Effect of EM Bokashi                              |              |                                     |                       |                              |                               |                                              |
| Interaction between plant extracts and EM Bokashi | control      | 4.03                                | 4.82                  | 4.96                         | 4.46                          | 4.57                                         |
|                                                   | EM Bokashi   | 5.71                                | 6.69                  | 7.08                         | 4.97                          | 6.11                                         |
| Effect of spraying with plant extracts            |              |                                     |                       |                              |                               |                                              |
| LSD 0.05                                          |              |                                     |                       |                              |                               |                                              |
| Cultivars                                         | EM Bokashi   | plant extracts                      | cultivars and Bokashi | cultivars and plant extracts | plant extracts and EM Bokashi | triple interaction                           |
| 0.61                                              | 0.50         | 0.71                                | 0.86                  | 1.20                         | 0.99                          | 1.73                                         |

The triple interaction treatment had a considerable influence on the proportion of total carbs in leaves. The maximum output was 11.41% in the treatment with the Summer Royal type of grape, 250 g of EM Bokashi, and moringa leaves. Meanwhile, the lowest productivity was 2.30% in the control treatment of the Crimson seedless cultivar with the control of EM and moringa petal extract treatments.

### Leaf auxin content

The auxin concentration in the leaves varies substantially across the grape cultivars (Table 4). The Summer Royal cultivar showed the largest rise in auxin content, with 16.52 mg of auxin per kg, while the Crimson cultivar had the lowest amount of auxin, with 12.93 mg of auxin per kg. The application of EM Bokashi to the soil boosted the soil's auxin content to a maximum of 15.33 mg kg<sup>-1</sup>, whereas the control had a high of 14.31 mg kg<sup>-1</sup>. The application of plant extracts to foliage raised the auxin content of plants: moringa leaves

produced 16.50 mg kg<sup>-1</sup>, and the lowest value of the control treatment was 13.63 mg kg<sup>-1</sup>.

Regarding the interaction between treatments, data on the same table showed significant variations between cultivars and the soil treatment process known as EM Bokashi. With a recorded value of 17.44 mg kg<sup>-1</sup>, the treatments (Summer Royal and EM Bokashi 250 g) had a notable impact, but the treatment (Crimson cultivar without additional components) had the lowest value, 12.55 mg kg<sup>-1</sup>. The leaves treated in the Summer Royal cultivar had the highest amount of auxin (20.69 mg kg<sup>-1</sup>) according to the interaction between cultivars and foliar application of plant extracts, whereas the leaves treated in the Crimson cultivar without spraying produced the lowest amount (12.55 mg kg<sup>-1</sup>). The concentration of auxin increased significantly when applying EM Bokashi and plant extracts topically. Meantime, the control treatment yielded a lower result of 12.89 mg/kg, and the treatment with 250 g of EM Bokashi and moringa leaf extracts gave a value of 17.19 mg/kg.

**Table 4.** Effect of Bokashi EM and foliar spraying with some plant extracts for three grape cultivars and their interactions on the leaf auxin content ( $\text{mg kg}^{-1}$ ).

| Cultivars                                         | EM Bokashi   | Foliar spraying with plant extracts |                       |                              |                               | Interaction between cultivars and Bokashi EM |
|---------------------------------------------------|--------------|-------------------------------------|-----------------------|------------------------------|-------------------------------|----------------------------------------------|
|                                                   |              | Control                             | Licorice root         | Moringa                      | Roselle petals/leaves         |                                              |
| Summer Royal seedless                             | control      | 11.92                               | 16.02                 | 19.53                        | 14.93                         | 15.60                                        |
|                                                   | EM Bokashi   | 15.37                               | 16.42                 | 21.84                        | 16.12                         | 17.44                                        |
| Flame seedless                                    | control      | 14.62                               | 14.84                 | 14.98                        | 14.75                         | 14.80                                        |
|                                                   | EM Bokashi   | 14.77                               | 15.02                 | 15.91                        | 15.16                         | 15.23                                        |
| Crimson seedless                                  | control      | 12.12                               | 12.68                 | 12.89                        | 12.42                         | 12.55                                        |
|                                                   | EM Bokashi   | 12.97                               | 13.35                 | 13.83                        | 13.22                         | 13.34                                        |
| Effect of cultivars                               |              |                                     |                       |                              |                               |                                              |
| Interaction between cultivars and plant extracts  | Summer Royal | 13.65                               | 16.22                 | 20.69                        | 15.52                         | 16.52                                        |
|                                                   | Flame        | 14.70                               | 14.93                 | 15.45                        | 14.96                         | 15.01                                        |
|                                                   | Crimson      | 12.55                               | 13.01                 | 13.36                        | 12.82                         | 12.93                                        |
| Effect of EM Bokashi                              |              |                                     |                       |                              |                               |                                              |
| Interaction between plant extracts and EM Bokashi | control      | 12.89                               | 14.51                 | 15.80                        | 14.03                         | 14.31                                        |
|                                                   | EM Bokashi   | 14.37                               | 14.93                 | 17.19                        | 14.83                         | 15.33                                        |
| Effect of spraying with plant extracts            |              |                                     |                       |                              |                               |                                              |
| LSD 0.05                                          |              |                                     |                       |                              |                               |                                              |
| Cultivars                                         | EM Bokashi   | plant extracts                      | cultivars and Bokashi | cultivars and plant extracts | plant extracts and EM Bokashi | triple interaction                           |
| 0.26                                              | 0.21         | 0.30                                | 0.36                  | 0.51                         | 0.42                          | 0.72                                         |

The triple treatment combination had a considerable influence on the auxin content in leaves. The treatment with the Summer Royal cultivar, whose weight is 250 g, and the moringa leaf extract had the heaviest value of 21.84 mg, whereas the control treatment had the lightest value of 11.92 mg.

### Leaf cytokinin content

The cytokinin content in the leaves of various grape cultivars was substantial (Table 5). The cultivar of Summer Royal showed a considerable rise in the level of cytokinins at  $14.05 \text{ mg kg}^{-1}$ , while the Crimson cultivar had the lowest content at  $8.90 \text{ mg kg}^{-1}$ . The application of EM Bokashi to the soil increased the content of cytokinins, with the maximum at  $12.13 \text{ mg kg}^{-1}$ , while the control had the minimum of  $10.91 \text{ mg kg}^{-1}$ . The leaves sprayed with extracts from plants attained a boost in the quantity of cytokinins: the moringa leaf extract had a yield of  $12.99 \text{ mg kg}^{-1}$ ; however, the control treatment had a minimum of  $10.57 \text{ mg kg}^{-1}$ .

The information in the same table revealed that the treatment plan sustained noteworthy influences from the interplay between cultivars and EM Bokashi soil application. With a yield of 14.8 g per square meter, the treatment on Summer Royal with the EM Bokashi (250 g) obtained a significant impact, whereas the Crimson treatment without additives had the lowest yield ( $8.61 \text{ g per square meter}$ ). The leaves treated on the Summer Royal cultivar had the highest amount of cytokinin in their foliage ( $17.36 \text{ mg kg}^{-1}$ ). Meanwhile, the leaves treated on the Crimson cultivar had the lowest amount ( $8.50 \text{ mg kg}^{-1}$ ), according to the interaction between cultivars and the foliar application of plant extracts. The quantities of cytokinins increased significantly when applying EM Bokashi and plant extracts topically. In contrast to the control treatment, which had a lower concentration of  $9.86 \text{ mg/kg}^{-1}$ , the treatment including 250 g of EM Bokashi and moringa leaf extract had a concentration of  $13.99 \text{ mg/kg}^{-1}$ .

The triple treatment combination had a considerable influence on the concentration of

**Table 5.** Effect of Bokashi EM and foliar spraying with some plant extracts for three grape cultivars and their interactions on the leaf cytokinin content (mg kg<sup>-1</sup>).

| Cultivars                                         | EM Bokashi   | Foliar spraying with plant extracts |                       |                              |                               | Interaction between cultivars and Bokashi EM |
|---------------------------------------------------|--------------|-------------------------------------|-----------------------|------------------------------|-------------------------------|----------------------------------------------|
|                                                   |              | Control                             | Licorice root         | Moringa                      | Roselle petals/leaves         |                                              |
| Summer Royal seedless                             | control      | 11.46                               | 13.92                 | 15.39                        | 12.44                         | 13.30                                        |
|                                                   | EM Bokashi   | 12.90                               | 13.67                 | 19.33                        | 13.29                         | 14.80                                        |
| Flame seedless                                    | control      | 10.04                               | 11.13                 | 11.59                        | 10.51                         | 10.82                                        |
|                                                   | EM Bokashi   | 12.02                               | 12.41                 | 13.03                        | 12.23                         | 12.42                                        |
| Crimson seedless                                  | control      | 8.08                                | 8.84                  | 9.01                         | 8.52                          | 8.61                                         |
|                                                   | EM Bokashi   | 8.93                                | 9.12                  | 9.64                         | 9.02                          | 9.18                                         |
| Effect of cultivars                               |              |                                     |                       |                              |                               |                                              |
| Interaction between cultivars and plant extracts  | Summer Royal | 12.18                               | 13.80                 | 17.36                        | 12.87                         | 14.05                                        |
|                                                   | Flame        | 11.03                               | 11.77                 | 12.31                        | 11.37                         | 11.62                                        |
|                                                   | Crimson      | 8.50                                | 8.98                  | 9.33                         | 8.77                          | 8.90                                         |
| Effect of EM Bokashi                              |              |                                     |                       |                              |                               |                                              |
| Interaction between plant extracts and EM Bokashi | control      | 9.86                                | 11.30                 | 11.99                        | 10.49                         | 10.91                                        |
|                                                   | EM Bokashi   | 11.28                               | 11.73                 | 13.99                        | 11.51                         | 12.13                                        |
| Effect of spraying with plant extracts            |              |                                     |                       |                              |                               |                                              |
| LSD 0.05                                          |              |                                     |                       |                              |                               |                                              |
| Cultivars                                         | EM Bokashi   | plant extracts                      | cultivars and Bokashi | cultivars and plant extracts | plant extracts and EM Bokashi | triple interaction                           |
| 0.19                                              | 0.16         | 0.23                                | 0.28                  | 0.39                         | 0.32                          | 0.55                                         |

cytokinins in leaves. The treatment with the Summer Royal cultivar, whose weight is 250 g, and the moringa leaf extract had the maximum value of 19.33 mg, whereas the control treatment had the minimum value of 8.08 mg.

## DISCUSSION

Earlier research revealed varied content of boron and total carbohydrate contents, as well as growth regulators (auxins and cytokinins) in the grape leaves. These advantages indicate relatively to the readily available assimilable nutrients. This availability helps the plant to access the nutrients it requires for development, particularly after the microorganisms in the growth media have digested the nutrients. This permits the plant to utilize the nutrients more effectively (Al-Khatib, 2007).

The EM Bokashi fertilizer boosts the activity of phosphatases in the soil and raises the biomass of microorganisms in the root zone. Theoretically, this enhances the phosphorus supply to the roots. The

microorganisms in EM Bokashi have a range of biological characteristics that enhance plant growth and boost the availability of critical components for growth and development. This improves the nutritional content of the plant (Bautista-Cruz *et al.*, 2014). This rise may also have associations with the physiological activity and the formation of nutritional balance in the seedlings, which has a good influence on vegetative development and the quantity of plant growth regulators (Prisa, 2020). Alternatively, this rise might be due to the influence of the EM Bokashi on soil fertility, which raises the efficiency of plant nutrient absorption and the production of growth hormones by microorganisms in the growth medium. Consequently, it leads to an increase in hormone concentrations in the plant. Moreover, the rise in leaf nutrient content improves the efficiency of photosynthesis and food processing, which has a positive impact on the studied characteristics (Japanese EMRO, 2003).

In the case of moringa leaf extract, the improvement in these characteristics of grape seedlings could be due to the extract

containing essential nutrients, especially nitrogen, phosphorus, and potassium, as well as micronutrients (Karthiga *et al.*, 2022). Nutrient intake is direct via the leaves, hence boosting the nutritional content in the plant. Another notion is that this extract could boost plant development by boosting nutrient absorption and photosynthesis, resulting in a greater nutritional content in the leaves (Abo-El-Enien *et al.*, 2015). The extract also supplies vitamins, minerals, phytochemicals, and amino acids and supports physiological activities, such as photosynthesis, respiration, chlorophyll creation, cell division, tissue development, and activation and control of plant hormones (Abdollahi *et al.*, 2011). Additionally, the extract includes vital minerals, i.e., iron, calcium, and potassium, wherein calcium and potassium play a significant role in plant development by regulating enzyme activity (Al-Hashimi, 2012).

## CONCLUSIONS

This investigation concludes the combined EM Bokashi (250 g of seedlings) soil application with the moringa leaf extract (15 ml/l) foliar spray and the seedless grape (*V. vinifera* L.) cultivar Summer Royal greatly affected the leaves' iron and boron concentrations. Likewise, such a combination influenced the total amount of carbohydrates and growth regulators (auxin and cytokinin) present in the soil and the growth of plants. As a result, the study advocates the treatments as they are natural and environmentally friendly substitutes for chemicals. Additionally, they have a remarkable role in enhancing the nutritional value of plants.

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