RESPONSE OF LOCAL SOUR ORANGE (CITRUS AURANTIUM L.) SEEDLINGS TO FOLIAR APPLICATION WITH TWO TYPES OF BIO STIMULATORS

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The study was conducted at the plastic house of the College of Agricultural Engineering Science, Salahaddin University - Erbil, Iraq, during 2022. To investigate the response of sour orange seedlings to application with bio health at three concentrations (0, 10 and 20 mg. L⁻¹) and three concentrations of Synergic (0, 3 and 6 ml. L⁻¹) on growth characteristics of Sour Orange (Citrus aurantium L.) seedlings local cultivar. A factorial experiment with three replicates was carried out in a Randomized Complete Design (CRD) using three seedlings for each experimental unit. Data were analyzed by (SAS) using the Duncan (ANOVA, P = 0.05). The obtained results indicated that the growth characteristics were increased by increasing the concentration of the two bio stimulators as compared with control, foliar application of bio health at (20 mg. L⁻¹) caused significant increase in most of the studied parameters. At the same time, foliar application of Synergic fertilization at (6 ml. L⁻¹) gave the highest value of stem high, stem diameter, leaf area, number of leaves /seedling, shoot fresh weight, shoot dry weight, root fresh weight and root dry weight. The interaction between the two bio stimulators caused a significant increase for all growth characteristics, the best treatment was at level (Bio. 20 mg. L⁻¹+ Sy. 6 ml.L⁻¹) as compare with control.

Keywords: Sour orange, Foliar application, Bio health, Synergic.
استجابة شتلات النارنج المحلي (Citrus aurantium L.) إلى الرش الورقي بنوعين من المحفزات الحيوية

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الخلاصة
أجريت الدراسة في البيت البلاستيكي التابع إلى كلية علوم الهندسة الزراعية، جامعة صلاح الدين - أربيل خلال عام 2022، لدراسة استجابة شتلات النارنج لرش الورقي من المحفز الحيوي Bio health بثلاثة تركيزات (0، 10 و20 ملغم لتر−1) وكذلك المحفز الحيوي Synergic بتثبيث تركيزات (0 و3 و6 مل. لتر−1) في صفات النمو الخضري لشتلات النارنج صنف محلة. نفذت تجربة عاملية ضمن تصميم تام التدفقات باستخدام ثلاث شتلات لكل وحدة تجريبية. تم تحويل النتائج بواسطة (SAS) باستخدام تحليل التباين (ANOVA، P < 0.05). أشارت النتائج المتحصل عليها إلى أن خصائص النمو ازدادت بزيادة تركيز المحفزين الحيويين قياسا بعاملة المقارنة. أدى الرش الورقي Bio health بتركيز 20 ملغم لتر−1 إلى زيادة معنوية في معظم الصفات المدروسة. في نفس الوقت، أعطى الرش الورقي بالمحفز Synergic عند تركيز 6 مل لتر−1 أعلى قيمة لكل من طول الساق، قطر الساق، مساحة الورقة، عدد الأوراق/ الشتلات، الوزن الطري للخضري، الوزن الجاف الخضري، الوزن الطري للجزر، والوزن الجاف للجزر. تسبب التداخل بين المحفزين الحيويين في زيادة معنوية لجميع خصائص النمو، وقد كانت أفضل النتائج عند المعاملة (6 مل لتر−1 + 20 ملغم لتر−1 من Bio health/Synergic).

كلمات مفتاحية: النارنج، الرش الورقي، Bio health، Synergic

Introduction

Citrus aurantium L. belongs to the citrus genus, which belongs to the Rutaceae family. Several studies claim that citrus originated in Southeast Asia, which is bordered by Northeast India, Burma, and Yunnan (China), but the most recent research suggests that citrus originated in Australia, New Caledonia, and New Guinea (11). Sweet orange, mandarin, sour/bitter orange, lemon, lime, grapefruit, and pomelos are examples of citrus plants (7) which important economically for edible fruits, drugs, aromatic leaves with oil glands, essential oil, and perfume. Citrus fruits contain flavonoids, which may have anticancer properties (10). They are also high in vitamin C, which is a powerful antioxidant that protects the body from free radical damage, and good sources of foliate and thiamin (13 and 14).
Citrus aurantium L., also known as Bitter orange, Seville orange, Bigarade orange, or Marmalade orange, is a tree. When used as a rootstock, roots have desirable characteristics, particularly for sweet orange scion cultivars (2). Because of its adequate yields, good fruit quality, tolerance to foot rot, cold, and compatibility with the most important Citrus species and varieties, Sour orange is the most widely used citrus rootstock (4). Plant-derived bio stimulants have been reported as a novel tool for addressing agricultural challenges and ensuring environmental sustainability. Furthermore, it has been reported that plant bio stimulants influence plant growth hormones, which improve plant metabolic activities and, as a result, crop productivity, chlorophyll synthesis, mineral status, and the synthesis and accumulation of antioxidant metabolites. These antioxidants boost plant growth by activating photosynthesis process (15).

Natural or synthetic substances that can be applied to seeds, plants, and soil are known as bio stimulants. These substances alter vital and structural processes in order to influence plant growth, increase seed and/or grain yield and quality, and improve tolerance to abiotic stresses. Furthermore, bio stimulants reduce the need for fertilizers (6).

Bio-fertilizers are microbial additives that contain bacterial or fungal cells, or both, and are added directly to seeds, plants, or soil to increase the readiness of plant nutrients and thus increase growth and production (16). Bio-fertilizers are extremely important in plant fertility and its surroundings because they work to provide one or more nutrients required for plant growth, reducing the need for mineral fertilizers while also encouraging plants to secrete some hormones and beneficial growth stimulants (1). These substances are effective in low concentrations, improving nutrition efficiency, abiotic stress tolerance, and/or crop quality traits regardless of nutrient content. When applied exogenously, these substances have actions similar to those of known plant hormones, the most important of which are auxins, gibberellins, and cytokinins (17).

In general, bio stimulants are created by combining natural or synthetic substances composed of hormones or plant hormone precursors. When used correctly in crops, it acts directly on physiological processes, potentially enhancing growth, development, and/or responses to water stress, saline, and toxic elements such as toxic aluminum. (3,5). In general, the highest values of vegetative growth parameters were obtained with potassium hamate, active dry yeast, and amino green in descending order; bio stimulants substances improved growth performance and leaf mineral content of Washington navel orange trees. (12).

Bio stimulants influence plant growth hormones, which improve plant metabolic activities and, as a result, crop productivity. They also improve chlorophyll synthesis, mineral status, and the synthesis and accumulation of antioxidant metabolites. These antioxidants boost plant growth by reactivating photosynthetic activity (15).
The objectives of this study were to evaluate the effects of spraying with the two-bio stimulator (Bio health and Synergiac) on some parameters growth of local sour orange seedlings to produce strong root system and to be ready for budding and grafting in next season.

Materials and Methods

The growth of local sour orange seedlings which planted in plastic pots (30 x 30 cm) 2 years old was studied in the plastic house/Horticulture department. The experiment included three concentration levels of Bio health and Synergetic by foliar application, as well as 0.1% Tween-20 as a wetting agent in all foliar solutions three times every 20 days. The study employs a completely randomized design (CRD) with three replications for each treatment. Duncan's multiple comparison tests were used to compare the treatments at the 5% level (SAS, 1996 and Al-Rawi and Khalafallah, 2000). Stem length, stem diameter, leaf area, number of leaves/seedling, shoot fresh weight, shoot dry weight, root fresh weight, and root dry weight were all measured for each treated seedling. The treatments that applied on sour orange seedlings, local cultivar; with three different levels namely:

1- Control, only sterilized water
2- Bio health 10 mg. L⁻¹.
3- Bio health 20 mg. L⁻¹.
4- Synergetic 3 ml. L⁻¹.
5- Synergetic 6 ml. L⁻¹.
6- Bio health 10 mg. L⁻¹ + Synergetic 3 ml. L⁻¹
7- Bio health 10 mg. L⁻¹ + Synergetic 6 ml. L⁻¹
8- Bio health 20 mg. L⁻¹ + Synergetic 3 ml. L⁻¹
9- Bio health 20 mg. L⁻¹ + Synergetic 6 ml. L⁻¹

Results and Discussion

It is clear from the data in table 1 that spraying Sour orange seedlings with the bio health at 20 mg. L⁻¹ concentration caused a significant promotion on growth parameters in terms of increasing stem length 31.03 cm, stem diameter 1.89 cm, leaf area 12.49 cm², number of leaves/shoot 18.61, shoot fresh weight 13.74 g, shoot dry weight 6.80 g, root fresh weight 10.39 g and root dry weight 5.25 g compared with the control treatment.

Table 1 Effect of bio health on some growth parameters of sour orange seedlings.

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Stem length cm</th>
<th>Stem diameter cm</th>
<th>Leaf area cm²</th>
<th>Number of Leaves/seedlings</th>
<th>Shoot fresh weight g</th>
<th>Shoot dry weight g</th>
<th>Root fresh weight g</th>
<th>Root dry weight g</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>18.94 c</td>
<td>0.73 c</td>
<td>6.90 c</td>
<td>11.55 c</td>
<td>8.94 c</td>
<td>3.86 c</td>
<td>6.01 c</td>
<td>3.02 c</td>
</tr>
<tr>
<td>10 mg. L⁻¹</td>
<td>26.87 b</td>
<td>1.60 b</td>
<td>8.76 b</td>
<td>15.23 b</td>
<td>11.21 b</td>
<td>5.45 b</td>
<td>8.43 b</td>
<td>4.15 b</td>
</tr>
<tr>
<td>20 mg. L⁻¹</td>
<td>31.03 a</td>
<td>1.89 a</td>
<td>12.49 a</td>
<td>18.61 a</td>
<td>13.74 a</td>
<td>6.80 a</td>
<td>10.39 a</td>
<td>5.25 a</td>
</tr>
</tbody>
</table>

*The similar letters vertically between treatments mean there are no significant differences between them using Duncan's Multiple Range test at P < 0.05.
It is evident from table 2 that all growth parameters were significantly affected by synergic treatment with concentration at level 6 ml. L⁻¹. Spraying Sour orange seedlings with synergic enhanced stem length 30.20 cm, stem diameter 1.90 cm, leaf area 11.82 cm², number of leaves/shoot 17.93, shoot fresh weight 12.53 g, shoot dry weight 6.37 g, root fresh weight 9.89 g and root dry weight 5.68 g compared with the control treatment.

Table 2 Effect of synergic on some growth parameters of sour orange seedlings.

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Stem length cm</th>
<th>Stem diameter cm</th>
<th>Leaf area cm²</th>
<th>Number of leaves/seedling</th>
<th>Shoot fresh weight g</th>
<th>Shoot dry weight g</th>
<th>Root fresh weight g</th>
<th>Root dry weight g</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>20.12 c</td>
<td>0.87 c</td>
<td>7.57 c</td>
<td>12.34 c</td>
<td>9.81 c</td>
<td>4.56 c</td>
<td>6.22 c</td>
<td>4.05 c</td>
</tr>
<tr>
<td>3 m. L⁻¹</td>
<td>27.94 b</td>
<td>1.54 b</td>
<td>8.94 b</td>
<td>15.87 b</td>
<td>10.77 b</td>
<td>5.61 b</td>
<td>7.48 b</td>
<td>4.90 b</td>
</tr>
<tr>
<td>6 m. L⁻¹</td>
<td>30.20 a</td>
<td>1.90 a</td>
<td>11.82 a</td>
<td>17.93 a</td>
<td>12.53 a</td>
<td>6.37 a</td>
<td>9.89 a</td>
<td>5.68 a</td>
</tr>
</tbody>
</table>

*The similar letters vertically between treatments mean there are no significant differences between them using Duncan's Multiple Range test at P < 0.05.

The data from table 3 shows the significant increase and recorded the highest value of all growth parameters at the interaction between bio health and synergic fertilizer at level (Bio 20 mg. L⁻¹ + Syn. 6 m. L⁻¹), even other interaction at level (Bio 10 mg. L⁻¹ + Syn. 6 m. L⁻¹) and (Bio 20 mg. L⁻¹ + Syn. 3 m. L⁻¹) had positive effect on the most growth parameters, stem length 33.87 cm, stem diameter 1.93 cm, leaf area 13.33 cm², number of leaves/shoot 20.09, shoot fresh weight 14.66 g, shoot dry weight 7.90 g, root fresh weight 12.20 g and root dry weight 6.33 g compared with the control treatment.

Table 3 Effect of interaction between bio health and synergic on some growth parameters of sour orange seedlings.

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Stem length cm</th>
<th>Stem diameter cm</th>
<th>Leaf area cm²</th>
<th>Number of leaves/seedling</th>
<th>Shoot fresh weight g</th>
<th>Shoot dry weight g</th>
<th>Root fresh weight g</th>
<th>Root dry weight g</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>15.14 h</td>
<td>0.55 g</td>
<td>5.48 h</td>
<td>8.10 h</td>
<td>7.52 f</td>
<td>3.22 d</td>
<td>8.12 f</td>
<td>3.29 e</td>
</tr>
<tr>
<td>0 + 10 mg. L⁻¹</td>
<td>18.98 g</td>
<td>0.67 f</td>
<td>7.11 g</td>
<td>14.33 f</td>
<td>8.77 e</td>
<td>3.47 d</td>
<td>8.30 e</td>
<td>3.55 e</td>
</tr>
<tr>
<td>0 + 20 mg. L⁻¹</td>
<td>23.86 e</td>
<td>1.21 e</td>
<td>8.09 f</td>
<td>15.35 e</td>
<td>10.40 c</td>
<td>5.02 bc</td>
<td>9.66 d</td>
<td>4.15 d</td>
</tr>
<tr>
<td>0 + 3 m. L⁻¹</td>
<td>22.39 f</td>
<td>0.72 d</td>
<td>9.54 e</td>
<td>13.28 g</td>
<td>9.87 d</td>
<td>4.11 c</td>
<td>9.76 d</td>
<td>4.21 d</td>
</tr>
<tr>
<td>0 + 6 m. L⁻¹</td>
<td>23.40 e</td>
<td>1.69 d</td>
<td>9.87 e</td>
<td>17.00 d</td>
<td>10.48 c</td>
<td>4.90 c</td>
<td>10.40 c</td>
<td>5.39 c</td>
</tr>
<tr>
<td>1 mg. L⁻¹ + 3 m. L⁻¹</td>
<td>25.81 d</td>
<td>1.77 ed</td>
<td>11.59 d</td>
<td>18.41 c</td>
<td>12.21 b</td>
<td>5.86 b</td>
<td>10.32 c</td>
<td>5.52 c</td>
</tr>
<tr>
<td>10mg.L⁻¹ + 6 m. L⁻¹</td>
<td>30.00 c</td>
<td>1.80 c</td>
<td>12.47 c</td>
<td>18.30 c</td>
<td>12.54 b</td>
<td>5.93 b</td>
<td>11.11 bc</td>
<td>5.19 c</td>
</tr>
<tr>
<td>20 mg.L⁻¹ + 3m. L⁻¹</td>
<td>31.29 b</td>
<td>1.87 b</td>
<td>13.02 b</td>
<td>19.32 b</td>
<td>14.09 a</td>
<td>7.01 a</td>
<td>11.79 b</td>
<td>6.00 ab</td>
</tr>
<tr>
<td>20mg.L⁻¹ + 6 m. L⁻¹</td>
<td>33.87 a</td>
<td>1.93 a</td>
<td>13.33 a</td>
<td>20.09 a</td>
<td>14.66 a</td>
<td>7.90 a</td>
<td>12.20 a</td>
<td>6.33 a</td>
</tr>
</tbody>
</table>

*The similar letters vertically between treatments mean there are no significant differences between them using Duncan's Multiple Range test at P < 0.05.

The top table results show that the true goal of this research was to determine the effect of different concentrations, application of bio stimulators on vegetative growth. The results showed that treatments with various concentrations of foliar bio stimulators had a positive effect on the vegetative parameters of sour orange
seedlings. A significant increase in Leaf area was observed, which resulted in an increase in photosynthesis, which resulted in a significant increase in Shoot fresh weight and Shoot dry weight. Increased root fresh weight and root dry weight, on the other hand, resulted in nutrient absorption from the soil and increased growth. The interaction of bio health, synergic, and concentrations in combination were also distinguished in the studied traits of the sour orange seedlings. Recently, a study on plant growth has been published. Plant growth stimulators have aided modern agriculture by improving plant growth and development, coping with biotic and abiotic stresses, and reducing heavy metal translocation (16).

Bio stimulants are nutrient-dense but cannot replace fertilizers. Even under stress conditions, it has the potential to improve soil quality and plant productivity. Bio stimulants are also responsible for increasing the chlorophyll content of the leaf. Its use on vegetables and floriculture crops has been shown to increase tolerance to biotic and abiotic stresses by improving internal and external quality (8). Bio stimulants influence plant growth hormones, which A. improve plant metabolic activities and, as a result, crop productivity (9). Furthermore, Bio stimulate is in charge of increasing leaf chlorophyll content and improving chlorophyll synthesis, mineral status, and the synthesis and accumulation of antioxidant metabolites. These antioxidants boost plant growth by reactivating photosynthetic activity (15).

Alternatively, it contains adequate quantities of nitrogen supply that increase the extra protein, allowing the plant foliage to grow larger and thus increasing its surface area available for photosynthesis, allowing the plants to grow faster, increasing the rate of metabolism, cell division, cell elongation, and thus stimulating apical growth as well as leaf formation (12). Bio-fertilizers are extremely important in plant fertility and its surroundings because they work to deliver nutrients required for plant growth, allowing mineral fertilizers to be reduced while also encouraging plants to excrete some hormones and beneficial growth stimulants (1). These findings are consistent with those of (6, 12, 15 and 17).

**Conclusion:** Plant growth stimulators have positively subsidized modern agriculture and have the potential to improve plant growth and development, as well as handle biotic and abiotic stresses. Bio stimulants are nutrient-dense but cannot replace fertilizers. Even under stress conditions, it has the potential to improve soil quality and plant productivity. There is a significant research gap. Different mechanisms, on the other hand, need to be studied further in order to develop a better understanding and introduce useful products

**Reference**


