EFFECT OF BIOPROMOTORS AND BIOFERTILIZERS APPLICATION ON GROWTH AND CHEMICAL COMPOSITION OF JOJOBA PLANTS

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ABSTRACT

A pot experiment was established during two successive seasons to evaluate the effect of biopromotor which is commercial product known as Biomagic (0.5,7,5,10g/l)and bacterial inoculation with Azotobacter, Bacillus and their mixture on the growth and chemical composition of jojoba (Simmondsia chinensis (Link) Schneider) plant. Experimental results indicated that, all applied treatments significantly increased vegetative growth parameters compared with control treatment. The highest values were always obtained from spraying plant with the high rate of Biomagic (10g/l) and inoculation with mixture of Azotobacter and Bacillus. The highest values of pigments were always obtained from spraying plant with the medium rate of Biomagic (7.5g/l) and inoculation with Azotobacter . The highest total carbohydrates in leaves, shoots and roots were recorded with spraying Biomagic at (10g/l) presence of mixture of Azotobacter, and Bacillus . The effect of Biomagic application and bacterial inoculation on leaves, shoots and roots content of mineral element was significantly increased compared with control treatment. Spraying Biomagic at high concentration (10g/l) plus inoculation with Azotobacter caused a high accumulation of N,K, Cu and Mn in jojoba leaves, shoots and roots followed by high concentration of Biomagic and inoculation with mixture of Azotobacter, and Bacillus . Using (10g/l) Biomagic and inoculation with Bacillus promoted a higher accumulation of elements P, Fe and Zn on jojoba tissues followed by the same concentration of Biomagic and dual inoculum.

INTRODUCTION

Jojoba { Simmondsia chinensis (Link) Schneider} pronounced hohoba, also called Goat Nut which belong to family, Simmondsiaceae, native to the southwestern United States, southern California, Arizona and northern Mexico (National Research Council,1985).

Jojoba is a perennial, dioecious, evergreen shrub or small tree that lives under diverse environmental conditions.

Recently, the use of extracts of living organisms as a foliar spray for the biostimulants has been increased . The most widely used biostimulants under the Egyptian conditions are that extracted from the micro-organisms, such as Biomagic, or from sea algae e.g. Promox, Sida compound, Dash or extraction of Baker’s yeast. Such extraction contains micro and macro elements, amino acid, vitamins, and plant hormones (El- Sayed, 2005).

Nitrogenous and phosphorus chemical fertilizers are commonly used, but with application of such fertilizer to the soil, some problems could be arise, e.g. some nitrogen could be lost via nitrate reduction, denitrification and / or ammonia volatilization. In addition, some amounts of nitrogen can be evaporated from soil surface and leached through under ground water, immobilization of phosphorus is the most important problem of phosphate
fertilization, which attributes to the high soil reaction where soil PH usually surpasses 8.0 causing inhibition in nutritional uptake. Therefore the use of biofertilizer is of particular interest to avoid the previously mentioned problems.

Moreover, microorganisms which are used as a biofertilizer induce stimulative effect of plant growth and production by fix atmospheric nitrogen in a free living state, e.g. Azotobacter and Azospirillum (Darwish, 2002).

In addition, Bacillus megatherium mobilize phosphate and micronutrients, while Azotobacter and Bacillus secret growth promoting factors, e.g. gibberellin, cytokinin like substances and auxins (Darwish, 2002).

The objective of this work is to examine the response of jojoba plant to Biomagic and bacterial inoculation under sandy soils with special emphasis on growth parameters and chemical composition.

MATERIALS AND METHODS

A pot experiment was carried out at the Experimental Nursery of Department of Ornamental Horticultural, Faculty of Agriculture, Cairo University, Giza during the period from 2002 until 2005. This investigation aimed to study the influence of foliar spray with biopromotor which is commercial product known as Biomagic (0.5, 7.5 and 10 g/l) and bacteria inoculation (uninoculated, Azotobacter chroococcum (Ar72) Bacillus megatherium var phosphaticum (Ar18), A. chroococcum xB. megatherium) on growth and chemical composition of jojoba plants (Simmondsia chinensis (Link) Schn.).

Seeds of jojoba were secured from El-kassasin Horticultural Experimental Station, Ismaillia Governorate, Ministry of Agriculture. On August 15th and 17th, 2002 and 2003 (in first and second season, respectively) the seeds of jojoba were sows in plastic pots (10 cm-diameter) filled with sandy soil.

On February 15th of each season the seedlings (20 cm, and 10-12 leaf/ plant in average) were planted in plastic pots (30 cm-diameter) filled with a constant weight (10 kg/each) of sandy soil. This soil was brought from El-Khatattba area, Menoufiya Governorate.

The physical characteristics of soil was carried out according to standard methods outlined by Klute (1986) and chemical analysis of soil was carried out according to standard methods outlined by Page (1982) and the data are listed in Table A.

All Plants received equal amount of chemical fertilizer and sheep manures. The chemical analysis of the organic fertilizer (Table, B) were made according to Page (1982).
### Table A. some physical and chemical properties of experimental soil samples.

<table>
<thead>
<tr>
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<tbody>
<tr>
<td>Ec mmhos/cm at 25°C</td>
<td>0.27</td>
<td>0.25</td>
</tr>
<tr>
<td>PH (1:2.5 H2O)</td>
<td>9.00</td>
<td>8.90</td>
</tr>
<tr>
<td><strong>Particle size distribution:</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sand</td>
<td>90.12</td>
<td>90.32</td>
</tr>
<tr>
<td>Silt</td>
<td>1.00</td>
<td>1.00</td>
</tr>
<tr>
<td>Clay</td>
<td>8.88</td>
<td>8.68</td>
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<tr>
<td><strong>Textural Class</strong></td>
<td>Sandy</td>
<td>Sandy</td>
</tr>
<tr>
<td><strong>Soluble ions (meq/1):</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Cations:</strong></td>
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</tr>
<tr>
<td>Ca$^{2+}$</td>
<td>0.20</td>
<td>0.60</td>
</tr>
<tr>
<td>Mg$^{2+}$</td>
<td>0.60</td>
<td>0.20</td>
</tr>
<tr>
<td>Na$^+$</td>
<td>1.14</td>
<td>1.11</td>
</tr>
<tr>
<td>K$^+$</td>
<td>0.22</td>
<td>0.26</td>
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<tr>
<td><strong>Anions:</strong></td>
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<td></td>
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<tr>
<td>CO$_3$$^{2-}$</td>
<td>--</td>
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<tr>
<td>HCO$_3$</td>
<td>0.60</td>
<td>0.80</td>
</tr>
<tr>
<td>Cl$^-$</td>
<td>1.50</td>
<td>1.00</td>
</tr>
<tr>
<td>SO$_4$$^{2-}$</td>
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<td><strong>Available nutrient (PPm):</strong></td>
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<td></td>
</tr>
<tr>
<td>N</td>
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<td>P</td>
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<td>K</td>
<td>32.00</td>
<td>80.00</td>
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<tr>
<td>Fe</td>
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<td>Cu</td>
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<td>Zn</td>
<td>0.10</td>
<td>0.16</td>
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<tr>
<td>Mn</td>
<td>0.80</td>
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<tbody>
<tr>
<td>Density (Kg/m³)</td>
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<tr>
<td>Relative humidity</td>
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<td>43.20</td>
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<td>Ec mmhos/cm at 25 °C</td>
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<td>PH (1:10 H2O)</td>
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<td>Organic mater</td>
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<tr>
<td>Organic Carbon</td>
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<td>28.1</td>
</tr>
<tr>
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<td></td>
</tr>
<tr>
<td>N (%)</td>
<td>1.68</td>
<td>1.51</td>
</tr>
<tr>
<td>P (%)</td>
<td>0.59</td>
<td>0.66</td>
</tr>
<tr>
<td>K (%)</td>
<td>4.78</td>
<td>5.98</td>
</tr>
<tr>
<td>Fe ppm</td>
<td>3310</td>
<td>3312</td>
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<tr>
<td>Zn ppm</td>
<td>94.80</td>
<td>133.05</td>
</tr>
<tr>
<td>Cu ppm</td>
<td>48.15</td>
<td>51.00</td>
</tr>
<tr>
<td>Mn ppm</td>
<td>264</td>
<td>274</td>
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</table>

All biofertilizer (Azotobacter chroococcum (Ar72) Bacillus megatherium var phosphaticum (Ar18), A. chroococcum xB. megatherium)
Sarhan, A. Z. et al

were added around the plant root in pots, the rate used were 150 ml/pot in each time on March and September in both seasons. Jojoba plants were sprayed with Biomagic periodically every 15 days starting from 15 days after transplanting (1st March) up to 31st October. Biomagic has PH of 5.5 and Consists of:

I- Amino acids (1.907%)

1. Arginine  
2. Cystine  
3. Glycine  
4. Histidine  
5. Isoleucine  
6. Leucine  
7. Lysine  
8. Phenylalanine  
9. Theronine
10. Tryptophane  
11. Tyrosine  
12. Valine

II- Vitamins (0.038%):

1. Thiamin  
2. Biotiene  
3. Choline  
4. Folic acid  
5. Niacin  
6. Pantothinic  
7. Pyrodxine  
8. Rhiboflavin

III- Macro elements (in mg/l):

1. N 1125  
2. P₂O₅ 550  
3. K₂O 625

IV. Micro elements (in mg/l):

1. Fe 160  
2. Zn 124  
3. Mn 100  
4. Mg 45  
5. Cu 45  
6. B  14  
7. Mo 12  
8. Cd  7  
9. Ni  4

The following data were recorded at the end of growth season (12 months).

- Plant height (cm)
- Stem diameter (cm)
- Number of branches / plant.
- Number of Leaves / plant.
- Fresh weight of leaves, shoots, and roots.
- Dry weight of leaves, shoots and roots.
- Pigments content (mg/g f. w.): chlorophyll a, b and carotenoids in the fresh leaves.
- Total carbohydrates contents percentage in the leaves, shoots and roots.
- Elements contents (N, P, K, Fe, Cu, Zn and Mn) were determined in the leaves, shoots and roots.

Chlorophylls and carotenoids were determined in leaf samples (mg/g fresh weight) using the method described by Nornai (1982). The Concentrations of total carbohydrates was determined in dried leaves, stem and roots using the methods described by Dubois et al. (1956).

Nitrogen concentration was determined by the micro kieldahl apparatus of Parnas Wanger as described Van-Schouwenburg and Walinga (1978). Phosphorus was estimated calorimetrically by using the chlorostannous reduced molybyphosphoric blue colour method as described by king (1951) .

Potassium concentration was determined by using the flame photometer (Corning 410).

Determination of micronutrients (Fe, Cu, Zn and Mn) concentrations were determined by the atomic absorption spectrophotometer (Thermo Jarrellas H, AA SCANI).
Experiments conducted in this study followed a Complete randomized design in factorial experiment. The obtained data were subjected to analysis of variance (ANOVA) according to Snedecor and Cochran (1980). Differences between means of treatments were compared using LSD values at 0.05 level.

RESULTS AND DISCUSSION

1- Vegetative growth characters

It is clear from data in Table 1,2 and 3 that Biomagic increased the different growth parameters compared with control. The stimulation effect of Biomagic in vegetative growth characters was increased with increasing the concentration used. The maximum values were recorded with the high concentration of Biomagic (10 g/l).

These results were in agreement with those obtained by Abdel-Hameed (2002) on olive trees, Aly (2002) on pea, Hanafy et al. (2007) on tomato, Salama (2005) on globe artichoke. Biomagic played an important regulatory role in plant growth development as well as yield and its component by enhancing the endogenous levels of various growth factors such as cytokines and gibberellins (Hanafy et al. 2007). It is worth here to mention that Biomagic is safe for environment to get lower chemical pollution effects. Also this natural compound reduced soil salinity via decrease using of mineral fertilization.

Regarding to the effect of bacterial inoculation, the obtained results indicate also that the alone inoculation with Azotobacter and Bacillus and their mixture, remarkably increased the different growth parameters compared with uninoculated plant as shown in the same tables. The least values were generally noticed with uninoculated control plant but the maximum values were recorded with treatment of mixture of Azotobacter and Bacillus.

These results are confirmed by Mohamed an Ram (1988) on Eucalyptus camaldulensis, Remesh et al. (1998) on nut seedlings, Abdel-Hameed (2002) on olive. Meantime it is clear that beside the primarily function of Azotobacter nitrogen fixation, it has been also well documented the ability of A. chroococcum to synthesize and secrete some vitamins such as thiamine, riboflavin, pyridoxine, cyanocobalamin, nicotinic and pantothenic acid, and some plant hormones e.g., indole substances, in addition to synthesizing antifungal antibiotics, which gave additional advantages for using A. chroococcum in the field or bio production (Subba Rao, 1993). Meanwhile, Bacillus megatherium has been found to convert bond phosphates, such as super phosphate and rock phosphate into forms, which are easily assimilated by the plant.

In general, data of vegetative growth of jojoba during the two seasons were significantly affected by application of Biomagic and dual inoculation with Azotobacter and Bacillus compared with the control and/or the most other treatments. It was also found that the maximum values were recorded with using the high rate of Biomagic (10g/l) when inoculation with mixture of Azotobacter and Bacillus.

6699
Sarhan, A. Z. et al

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T2
Sarhan, A. Z. et al

T3

6702
II- Pigments :

1- Chlorophyll "a" content (mg/g f.w.) :

It was clear from data in Table (4) for both seasons that chlorophyll "a" content of jojoba plant was considerably influenced by spraying with Biomagic. Application of Biomagic at different rates led to increase in the leaf content of chlorophyll "a" compared with untreated plants. Control plants had the lowest chlorophyll a content. Meanwhile Biomagic at the medium rate (7.5 g/l) produced the highest value of chlorophyll "a" content followed by the high rate.

Regarding the effect of bacteria on the chlorophyll "a" content of jojoba leaves. The least values were generally noticed with uninoculated control plants during two seasons. The highest chlorophyll "a" content was recorded with the plants inoculated with Azotobacter followed by the treatment of inoculation with the two bacterial mixture.

Moreover, the interaction effect between Biomagic rates and bacterial inoculation on the chlorophyll "a" content of jojoba in Table (4) data reveal that, the highest value of percentage content was produced with sprayed plants with Biomagic at the rate of 7.5 g/l when inoculated with Azotobacter. In the second rank lies the value observed with plants sprayed with Biomagic at the rate of 7.5 g/l when inoculated with mixture of bacteria. The increases in chlorophyll "a" content with these two treatments were significant when compared with the control. Similar results was recorded by Sharma and Bhutani (1998) on apple seedlings.

2- Chlorophyll "b" content (mg/g f.w.) :

The results in Table (4) indicate that the chlorophyll "b" content followed a trend similar to that obtained in the chlorophyll "a". The medium and high rates of Biomagic showed a more pronounced effect on chlorophyll "b" content than untreated or the low rate of Biomagic.

Also, the Chlorophyll "b" content was considerably affected by inoculation with bacteria. The highest chlorophyll "b" content was recorded with plants inoculated with Azotobacter followed by the treatment of inoculation with the two bacterial mixture, but without significance between themselves.

Data presented in Table (4) showed that chlorophyll "b" content affected by the combination between Biomagic and bacteria treatments in the first season. The treatments which included Biomagic at the rate of 7.5 g/l and inoculation with Azotobacter alone and the mixture of Azotobacter + Bacillus were more effective and show increasing on the synthesis and accumulation of chlorophyll "b" than control and the other treatments. On the other hand in the second season treated plant with high Biomagic concentration (10g/l) and dual inoculation of Azotobacter and Bacillus produced the best values of chlorophyll "b".
Sarhan, A. Z. et al

T4
3- Total chlorophylls (a+b) (mg/g f.w.) :
Regarding the effect of different Biomagic concentrations on the leaves total chlorophylls (a+b) (Table4) it noticed that, all Biomagic treatments had a pronounced effect on increasing the content of total chlorophylls in leaves compared to control. It can observed that Biomagic at medium concentration (7.5 g/l) had considerable effect on the leaf content and gave the highest percentage of total chlorophylls in leaves.

It can be noticed that the total chlorophylls (a+b) content was considerably affected by inoculation with bacteria. The highest total chlorophylls content was recorded with plants inoculated with Azotobacter followed by the treatment of inoculation with the two bacterial mixture, but without significance between themselves.

As for the combination effect of Biomagic rates and bacterial inoculation on the total chlorophylls of jojoba leaves, the highest of percentage content was produced with sprayed plants with Biomagic at the rate of (7.5 g/l) when inoculated with Azotobacter.

Carotenoides content (mg/g f.w.)
The data in Table (4) indicate that, application of Biomagic at different rates led to increase in the leaf content of carotenoides compared with control plants. Sprayed plants with Biomagic at (10 g/l) produced the highest value of carotenoides content.

Regarding the effect of bacterial inoculation Table (4) on the carotenoids content of jojoba leaves, uninoculated control had lower carotenoids content than inoculated. The highest carotenoids content determined in leaves of plants inoculated with mixture of bacteria in the first season and Azotobacter in the second season.

Data presented in Table (4) showed that carotenoides content was positively affected by either increasing Biomagic rates or Bacteria inoculation. The best values were recorded with using the 10g/l Biomagic with individual Azotobacter followed by 7.5 g/l Biomagic with inoculation by Azotobacter alone or mixed with Bacillus but without signficancy between themselves. Similar results was recorded by Aly (2002) on pea plants.

III Total carbohydrates (%):
Experimental data presented in Table (5) indicated that the total carbohydrates of leaves, shoots and roots were significantly affected by Biomagic rates. The general increase of total carbohydrates in leaves,shoots and roots as a result of using Biomagic treatments are shown in Table (5). Plant supplied with different Biomagic concentration recorded an increase in total carbohydrates in their leaves, shoots and roots. The greatest values were recorded on plant treated by the high Biomagic rate (10g/l), and followed by the medium rate (7.5g/l), but without signficancy between themselves.

Concerning the effect of bacterial inoculation in general, the values of total carbohydrates in leaves, shoots and roots were increased due to inoculation treatments.
Sarhan, A. Z. et al

T5
In this regard, the least values were generally noticed with the uninoculated control plant during two seasons but the best values were recorded with dual inoculation treatment. As for the combination effect of Biomagic rates and bacterial inoculation on the total carbohydrates in leaves, shoots and roots. The highest total carbohydrates in leaves, shoots and roots were recorded with plants inoculated with Azotobacter and Bacillus presence of high concentration of Biomagic (10 g/l).

V. Nutrient contents:

1. Nitrogen Content (%):

Data presented in Table (6) reflected significant effect for Biomagic concentrations on nitrogen contents in leaves, shoots and roots. Using Biomagic at high rate (10 g/l) markedly promoted a high nitrogen content in leaves, shoots and roots of jojoba plants compared to control and other rates of Biomagic.

Bacterial inoculation, in most cases, reflected enhancement of N content of leaves, shoots and roots compared to the uninoculated treatment, especially in case of Azotobacter where the highest values were recorded followed by dual inoculation treatment but without significancy between themselves.

Nitrogen uptake by leave, shoot and root of jojoba plants was presented in Table (6). It seems that the N uptake was positively affected by Biomagic and bacteria inoculation. The highest N content recorded with treatment of spraying with Biomagic at the high concentration (10 g/l) and inoculation with Azotobacter, followed by the treatment of inoculation with the two bacterial mixture amended with high rate of Biomagic (10 g/l) but without significany between them.

2- Phosphorus content (%):

It is clear from data in table (7) that P percentages in leaves, shoots and roots of jojoba plants were considerably increased by Biomagic treatments. The highest P% recorded with the treatment of high concentration of Biomagic (10g/l). The favorable effects of Biomagic treatments on P- content on jojoba were similar to that result obtained by Abdel- Hameed (2002) on olive, Aly (2002) on Pea, Salama (2005) on globe artichoke.

The obtained results indicate also that the alone inoculation with Azotobacter or Bacillus, and their mixture slightly increased the P content compared with uninoculated ones. The highest P percentages in leaves, shoots and roots of jojoba plant were recorded with the treatment of Bacillus inoculation followed by dual inoculation of Azotobacter and Bacillus, but without significancy between themselves. Similar results were recorded by, Sorial et al. (1992) on tomato, Haggag et. al (1994) guava seedlings.

As regards, the combination effect of Biomagic rates and bacterial inoculation on concentration of P in leaves, shoots and roots of jojoba plant data revealed that the highest P% recorded with the treatment of Biomagic at the high concentration (10g/l) with Bacillus inoculation followed by above concentration of Biomagic (10 g/l) with the treatment of dual inoculation of Azotobacter and Bacillus.
Sarhan, A. Z. et al

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3- Potassium content (%):

It is evident from the results in Table (8) that K-content showed similar trend as that in N content. The different Biomagic concentrations had generally a little effect on K- contents in leaves, shoots and roots of jojoba plant.

Concerning the effect of bacterial inoculation, it was found that the maximum values were recorded with the treatment of inoculation with *Azotobacter* followed by mixture of *Azotobacter and Bacillus*, but without significance between themselves. Meantime, it is clear that spraying Biomagic with (10g/l) to *Azotobacter* and/or mixture of *Azotobacter and Bacillus* generally increased the obtained results over the control.

4- Iron content (mg/g):

Data presented in Table (9) showed that Fe uptake by leaves, shoots and roots was positively affected by either increasing Biomagic concentration or bacterial inoculation.

Concerning the effect of Biomagic, it is obvious that Fe absorption was gradually increased with increasing Biomagic concentration. Also, the results of both leaves, shoots and roots were nearly closed to each other. Generally, the highest values of Fe content in leaves, shoots or roots were recorded with spraying high concentration of Biomagic (10g/l). Similar result, was recorded by Abdel – Hameed (2002) on olive.

From the results, it can be recognized that, bacterial inoculation (Table,9 ) increased Fe uptake by leaves, shoots, or roots especially in case of *Azotobacter* inoculation treatment where, the highest values were recorded, followed by dual inoculation treatment, but without significance between them. The Fe contents as affected by the interaction effect between Biomagic and bacteria. The highest value of Fe was produced with plants inoculated with Bacillus or Azotobacter and Bacillus, in the presence of spraying with (10g/l) Biomagic.

5- Copper Content (mg/g):

It is evident from the data in Table (10) that Biomagic treatments considerably affected the Cu contents in leaves, shoots and roost of jojoba plants, i.e. sprayed plants had more Cu concentration in their tissues than control plants. The more effective concentration was Biomagic at (10g/l).

Concerning the effect of bacterial inoculation, in general, increase in Cu uptake by leaves, shoots, and roots especially in case of *Azotobacter* inoculation treatment where the highest values were recorded.

As for effect of interaction between applied Biomagic and bacterial inoculation in the (Table 10), the combination between high concentration of Biomagic (10g/l) and *Azotobacter* inoculation was more effective in increasing Cu content in jojoba tissues than other combination inoculation. In general, increase in Cu uptake by leaves, shoots, and roots especially in case of *Azotobacter* inoculation treatment where the highest values were recorded.
6711
Sarhan, A. Z. et al

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As for effect of interaction between applied Biomagic and bacterial inoculation in the (Table 10), the combination between high concentration of Biomagic (10g/l) and Azotobacter inoculation was more effective in increasing Cu content in jojoba tissues than other combination.

6- Zinc content (mg/g):

As shown in Table (11), the acquisition of Zn by leaves, shoots and roots of jojoba plants was frequently affected by either Biomagic or bacterial inoculation. Zinc in leaves, shoots and roots tended to increase with increasing Biomagic. Thus the highest content of Zn content was recorded on plants sprayed with Biomagic at the concentration of (10g/l).

Concerning the effect of bacterial inoculation in general, the values of Zn uptake by leaves, shoots and roots were increased by inoculation treatments. In this regard the best values were recorded with individual Bacillus followed by the treatment of inoculation with the two bacterial mixture. Regarding the effect of Biomagic rates combined with bacterial inoculation treatments on the Zn content Table (11), data reveal that, this combination had pronounced effect an increasing the content of Zn than control plants.

Generally, the highest values of Zn content in leaves, shoots or roots were recorded by spraying with high concentration of Biomagic and inoculation with Bacillus.

7- Manganese content:

Data in Table (12) reveal that, Biomagic treatments had remarkable effect on Mn content in leaves, shoots and roots of jojoba plant. The highest Mn content was produced with plants sprayed with Biomagic at high concentration (10g/l). It appears from Table (12) that the least values of Mn in leaves, shoots and roots were generally noticed with the uninoculated control plant during the two seasons. The highest Mn content was produced with plants inoculated with individual Azotobacter followed by dual inoculum but without significancy between themselves.

As for the effect of interaction between Biomagic treatments and bacterial inoculation, in general the best values were recorded in leaves, shoots and roots of jojoba plants were recorded with spraying at the high rate of Biomagic (10g/l) combined with individual Azotobacter followed by using the high rate of Biomagic (10g/l) and inoculation with two bacterial mixture, but without significancy between themselves.
REFERENCES


6717


Sarhan, A. Z. et al

6718