Effect of chemical and organic fertilization and 
Aztobacter chorocoocum inoculation on the 
bacterial biomass and yield of tomato plant 
(Lycopersicon esculentum Mill) 

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Abstract

The tomato crop (Bride of Spring) 2019 was planted in a field within the Thurima project Al-Qadisiyah Governorate, and the mineral NPK fertilizer was used in three levels, organic fertilizer (composed poultry) with three levels and a biological fertilizer consisting of Aztobacter chorocoocum bacteria after that. Isolation and diagnosis of it on two levels. The results of the treatment of half the recommendation of compost in combination with the third level of organic fertilizer and bio-fertilizer gave results that do not differ significantly from the treatment of the recommendation of the full fertilizer in the above triple interaction in soil characteristics and plants (Total Bactiria, The productivity of one plant) This indicates the importance of the role of organic fertilizer Bio-reduction in mineral fertilizers.

Keywords: Aztobacter chorocoocum, mineral fertilizer and organic fertilizer.

1. Introduction

The increase in the amount of mineral fertilizer added leads to an increase in the readiness of phosphorus in the soil that can be absorbed by the roots, which in turn reflected on the concentration of phosphorus in the plant, as phosphorus strengthens and activates the root system of plants and thus increases the absorption of nutrients and water for the plant [1]. Azotobacter also plays an important role in increasing soil nitrogen, as well as its ability to secrete stimulants and growth regulators, which led to increased root system growth and increased absorption of nutrients in the plant [2,3]. The use of organic fertilizers in agriculture increases the soil's content of organic matter, improves its physical and chemical properties, and encourages micro-organism activity, thus improving fertile soil properties and productivity. Therefore, the world has recently turned to the use of modern fertilizer technologies to reduce pollution problems and interest in the technology of bio-organic agriculture, in which organic fertilizers and beneficial microorganisms are used in order to improve agricultural production in quantity and quality [4,5].

This improvement and increase can be attributed to the role of biological fertilization in improving the level of nitrogen and phosphorous in the plant as a result of fixing nitrogen and dissolving phosphorous and improving plant growth through the secretion of growth regulators, the most important of which are auxins, gibberellins and cytokines that encourage plant absorption of nutrients, including potassium [6]. As [7], showed that organic and biological fertilizers can reduce the quantities of mineral fertilizers if they are used together. The reason is attributed to the role of organic matter if it works to preserve PH, which increases phosphorous readiness [8,9].

Studies have confirmed the role of organic matter in activating Azotobacter and thus increasing their effective numbers in fixing atmospheric nitrogen and increasing secretion of photo hormones by providing the bacteria with energy that is a prerequisite for reproduction. Better results and reduce as much as possible the use of mineral fertilizers [7,10].
2. Materials and methods

Preparation of bacterial biofertilizers: 20 soil samples were collected from different fields cultivated with tomato crops, cucumbers, watermelon, and eggplant. Samples were taken from the rhizosphere soil of crop roots to different locations within the same field and from a depth of 0-30 cm and mixed to form a compound sample and packed in clean and sterile polyethylene bags and transported to the laboratory. Physical, chemical and biological analyzes were performed as shown in Table (1) and kept in the refrigerator before use In the process of isolation.

| Unit        | Value   | Property          |
|-------------|---------|-------------------|
| Clay, gm.kg.soil^-1 | 331     | Clay              |
| Silt, gm.kg.soil^-1   | 255     | Sand              |
| Sand, gm.kg.soil^-1   | 414     |                   |
| Clay mixtures gm.cm^-3 | 1.37    | Texture type      |
| d. Sm^-1             | 7.55    | pH(1:1)           |
| d. Sm^-1             | 2.36    | EC, Electrical conductivity |
| Cante moul.kg^-1 soil gm.kg.soil^-1 | 36      | CEC Cation exchange Capacity |
| O.M Organic matter gm.kg.soil^-1 | 20.10   | Nitrogen          |
| phosphorous gm.kg.soil^-1 | 3.34    | Aions avilable    |
| Potassium gm.kg.soil^-1 | 138.95  | Calcium           |
| Calcium C.mol L^-1    | 1.83    | magnesium         |
| Sodium C.mol L^-1     | 3.32    | Cationic soluble ions |
| Nill C.mol L^-1       | 2.51    | Carbonate         |
| Bicarbonate C.mol L^-1| 1.40    | Negative soluble ions |
| Sulfates              | 4.01*10^-10 | Total bacteria     |
| CFU gm^-1 dry soil    | 1.54 * 10^7 | Biological Estimates |

2.1 Isolation of Azotobacter spp

Soil loosening was prepared by adding 10 g of the selected samples to 90 ml of sterile distilled water in 250-capacity flasks of liquid medium (Sucrose mineral-salts) and the test tubes were placed in the incubator at a temperature of 28 °C for 3 days and the tubes were examined by observing the brown membrane (brown ring) Which is an indicator of the growth of Azotobacter spp. Use 0.1 ml of dilution, which gave an indication of the growth of Azotobacter spp., and spread it in a plate containing the solid medium Sucrose mineral-salt with 2% agglutinative addition and with three repetitions. The dishes were incubated for 48 hours and then films were prepared from them and it was stained by a Gram method and examined microscopically by using light microscopy to study the phenotypic characteristics. 10 pure isolates of A. chroococcum were obtained.

2.2 Diagnosis of Azotobacter spp

To ensure the purity of the culture and the success of the isolation process, membranes from the ten isolates diagonal farms were prepared on 48 hour old concave glass slides and stained in the Gram method. The shapes of cells, their size, and the ability of bacteria to interact with the dye were studied using the oily lens [11].
2.3. Morphological characteristics

The morphological (phenotypic) characteristics of *A. chroococcum* isolates were studied by observing the bacterial growth on the solid culture media inside the plates and studying the bacterial colony's shape, height, edge shape, surface, size, transparency, texture, and the amount of bacterial growth. And the production of pigments according to the method [11,12].

2.4. Bio-tests for bacteria

The following tests were conducted, growth at a temperature of 37°C, starch decomposition test, amid nitrate reduction, Catalase test, oxidase test, fermentation test of sugars amid movement test, efficiency test of isolates on atmospheric nitrogen fixation and the amount of ammonia was estimated It was formed in the middle by taking 2 ml of it and assessing it with a Microkildahl device (2 and 1) and isolate No. (1) (A1) the quantity of N2 fixed (mg. L⁻¹), was selected [13]. After contaminating the tomato seeds with using gum arabic after removing the sterile substance (4 and 3).

2.5. Field measurements

By the yield of one plant (kg. Plant⁻¹) and the number of total bacteria was calculated according to the method of greatest probability (MPN).

2.6. Preparing the soil for the greenhouse

The experiment was carried out in a plastic house with dimensions of (9 * 52) m in mixed clay soil with certain physical, chemical and biological properties. The soil was prepared and smoothed well, soil samples were taken as representative as possible from the surface layer (0-30 cm) Then it was dried and milled with a polyethylene hammer and passed through a sieve with a diameter of 2 mm holes and mixed well and stored samples of it in the refrigerator at a temperature of 4°C for biological analyzes. Three floors were established along the plastic house between each floor and the other, an isolation distance of 2 meters and divided one into 16 units. Experimental length of one is 2.5 m and a width of 1.25 m, including an isolation distance of 0.75 m to prevent interferences between experimental units in the same sector [14,15], and the fertilizer recommendation was added according to the recommendations of the Iraqi Ministry of Agriculture.

2.7. Experimental design

An Experimental factorial was carried out according to R.C.B.D, with three replications. The experiment included [2] treatment as in the following :-

- Fertilizer recommendation added 320-320-160 t. h⁻¹ (N-P₂O₅-K₂O) and in three levels (0, 50, 100)% of it.
- Levels of *Azotobacter chroococcum* bacterial inoculation (A₀ without inoculation and A₁ inoculation).
- Levels of organic fertilizer (2, 1, 0) t. h⁻¹.

3. Results and discussion

Table (2) indicated that mineral fertilization at the level of 100% NPK gave an increase in the number of bacteria, if the value of the treatment 9.14 cfu gave a significant increase in comparison with the control treatment, and the reason was attributed to the increase in nutrients that lead to an increase in the bacterial community [16]. The results are consistent with [17]. The table also shows that the addition of the organic matter in the third level gave an increase in the number of bacteria, as the value of the treatment 8.10 cuf gave a significant increase compared to the control treatment, and the reason is due to the fact that the organic matter contains many of the nutrients that the microscopic organisms use in building their bodies 18 and agree These results are with [18]. The inoculation with ectopic bacteria also increased the number of bacteria, as the inoculation treatment achieved a significant increase in the number of bacteria in the treatment 8.31 cuf compared to the control treatment. Results with [19,20]. As for the bilateral overlap, the table indicated that the numbers of total bacteria increased in all levels of organic matter, with levels of mineral fertilizer, the highest achieved was a value of 9.04 cuf in comparison with the control treatment. The reason is due to the availability of important nutrients such as NPK, sulfur, and others, which are of great importance in life Microorganism (34 and 28). These results are in agreement with [21].

The table also showed that the two-way interaction between mineral and biological fertilizers led to an increase in the number of total bacteria, the highest of which was when the treatment value was 8.31 cuf The reason was attributed to the importance of nutrients in the diet of microorganisms and the results are consistent with [17,22]. The table showed the role of the interaction between organic and biological fertilizers in increasing the number of total bacteria in the soil of the rhizosphere, as all levels of organic fertilizer achieved a significant increase in the number of bacteria, the highest in the treatment value
being 9.70 cuf compared to the control treatment. The reason for this is attributed to the role of organic matter and it contains important nutrients in large proportions in feeding micro-organisms as well as the role of nutrients in mineral fertilizers [4], and these results agree with [23,24]. The triple interdiction (mineral, organic and biological fertilizers) gave a significant increase in the value of the treatment 7.79 cuf compared with the organic matter, indicating that the second level of fertilizer recommendation is 50% NPK with the third level of organic matter.

Table 2. The effect of chemical, organic and biological fertilization on the numbers of total bacteria.

| Interaction M*O | Levels of bio-fertilization B | Without inoculation | Fertilization levels Organic O | Fertilization levels M |
|-----------------|-------------------------------|---------------------|--------------------------------|-----------------------|
| 2.99            | 3.68                          | 2.95                | 0                              | NPK 0 %               |
| 3.99            | 4.57                          | 3.97                | 1                              | NPK 50 %              |
| 5.25            | .226                          | 5.15                | 2                              |                       |
| 6.60            | 6.76                          | 6.50                | 0                              |                       |
| 7.00            | 7.00                          | 7.00                | 1                              |                       |
| 7.78            | 7.79                          | 7.68                | 2                              |                       |
| 8.20            | 8.31                          | 8.10                | 0                              |                       |
| 8.75            | 8.89                          | 8.65                | 1                              | NPK 100 %             |
| 9.14            | 9.70                          | 9.04                | 2                              |                       |

L.s.d 0.05 0.29

Level M

| 2.97            | 3.99                          | 2.95                | NPK 0 %                        |
| 6.60            | 6.76                          | 6.50                | NPK 50 %                       |
| 9.14            | 8.31                          | 8.10                | NPK 100 %                      |

L.s.d 0.05 0.86

Level O

| 3.01            | 7.97                          | 2.96                | 0                              |
| 6.50            | 6.76                          | 8.30                | 1                              |
| 8.10            | 8.31                          | 7.23                | 2                              |

L.s.d 0.05 0.79

L.s.d 0.05 0.82

Total bio-fertilizer levels

Table (3) indicated that mineral fertilization at the level of 100% NPK gave an increase in tomato plant yield (kg. Plant -1), if the treatment value gave (kg. Plant -1) a significant increase compared to the control treatment, and the reason is attributed to that The increase in nutrients leads to an increase in the nutrient in the plant, which leads to an increase in all the various plant activities [18], and the results are consistent with [3,21]. The table also shows that the addition of the organic matter in the third level gave an increase, as the value of the treatment (kg. Plant -1) gave an increase in the yield of the tomato plant compared to the control treatment, and the reason is that the organic matter contains many nutrients that the plant benefits from Of which the growth of which is S, NPK [25], and these results are consistent with [4,23]. Also, the inoculation with Aztobacter increased the number of bacteria, as the pollination treatment achieved a significant increase in the yield of tomato plant (kg. Plant-1), compared to the control treatment. Increase the branching of the roots and thus increase the material absorbed in the plant, which is reflected in its overall vital activities [26], and these results agree with [5,27].

As for the bilateral interaction of organic matter with mineral fertilizer, the table indicated that an increase in tomato plant yield (kg. Plant -1) increased in all levels of organic matter with levels of mineral fertilizer, the highest was achieved value (kg. Plant -1) in comparison with the treatment of The control is due to the availability of important nutrients such as NPK, sulfur and others, which are of great importance in plant life and production [13], and these results are consistent with [16,21]. The table also showed that the two-way interaction between mineral and biological fertilizers led to an increase in the yield of tomato plant (kg. Plant -1), the highest was when the treatment value (kg. Plant -1). The reason is due to the importance of the nutrients NPK and the rest of the elements contained in the organic matter. And minerals in the plant's diet [23], and the results are consistent with [5,28].
The table showed the role of the interaction between organic and biological fertilizers in, as all levels of organic fertilizer achieved an increase in, as the value of the treatment was achieved (kg. Plant⁻¹) in comparison with the control treatment. The reason for this is attributed to the role of organic matter and it contains important nutrients in large proportions in plant nutrition, as well as the role of nutrients in mineral fertilizers, which is reflected in flowering processes and the size of fruits [29], and these results agree with [3,18]. As for the triple overlap (mineral, organic and biological fertilizers), there was an increase in the yield of tomato plant (kg. Plant⁻¹), if the value of the treatment (cm. Plant⁻¹) gave a significant increase in comparison with the control treatment, indicating that the second level of recommendation Fertilizer 50% NPK with the third level of organic matter achieved a significant increase and thus led to a decrease in mineral fertilizers.

Table 3. The effect of chemical, organic and biological fertilization on tomato plant yield in( kg.plant⁻¹).

| Interaction | Levels of bio-fertilization | Fertilization levels | Fertilization levels |
|-------------|-----------------------------|----------------------|----------------------|
| M*O         | Without inoculation         | Organic O            | NPK 0 %              |
| 5.50        | 5.62                        | 5.53                 | 0                    |
| 5.65        | 5.86                        | 5.78                 | 1                    |
| 6.36        | 6.13                        | 6.46                 | 2                    |
| 5.85        | 6.77                        | 5.95                 | 0                    |
| 6.11        | 6.38                        | 6.21                 | 1                    |
| 6.66        | 6.72                        | 6.45                 | 2                    |
| 6.33        | 6.43                        | 8.43                 | 0                    |
| 6.58        | 6.68                        | 6.68                 | 1                    |
| 6.76        | 6.86                        | 6.76                 | 2                    |
| L.s.d 0.05  | 0.15                        | L.s.d 0.05           | 0.002                |
| M          | levels                      |                      |                      |
| 5.53        | 5.78                        | 5.53                 | NPK 0 %              |
| 5.75        | 6.77                        | 5.95                 | NPK 50 %             |
| 5.84        | 6.79                        | 5.84                 | NPK 100 %            |
| L.s.d 0.05  | 0.14                        | L.s.d 0.05           | 0.25                 |

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