The Effect of Bacterial R. liguminosarum and Fungal T. harzinum Inoculation and Organic Manure on Root Nodes, Growth, and Yield of Mung Bean, Vigna radiate

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Abstract

This study aimed to investigate the effect of the bacterial inoculum R. liguminosarum, T. harzinum, levels of organic fertilizer (0, 5, and 10 t ha⁻¹), and their overlap the root nodes (number of root nodes and weight of root nodes) and the growth and yield of a local variety of mung bean (Dry weight of root mass, leaf chlorophyll content and grain yield). A field experiment was conducted in the fall season of 2020 in one of the fields of Al-Diwaniyah Governorate / Al-Daghara county. The experiment was carried out according to the Randomized Complete Block Design (R.C.B.D) with three replications. The results of the statistical analysis of the least significant difference (L.S.D.) present that the overlapping treatment (bacterial inoculum, fungal inoculum, and five t ha⁻¹ organic fertilizer) resulted in the highest means: number of root nodes, root nodule weight, dry weight of root total, chlorophyll content in leaves and grain yield, (72.33) Knots.plants⁻¹, (463) gm Plants⁻¹, (1,547) gm Plants⁻¹, (59.14) SPAD, and (3,531) t ha⁻¹, respectively, by comparison with comparison treatment.

Keywords: R.leguminosarum, T.harzinum, compost, root nodes, mung bean.

1. Introduction

Mung bean, Vigna radiate L., belongs to the Leguminosae family. It is one of the essential food legumes cultivated worldwide and originated in the Indian subcontinent. It is one of the most common crops in most tropical and subtropical regions. It has a short growing season (90-120) days and tolerates drought and heat conditions in all growth stages except for the flowering stage [1]. Mung bean is usually grown to obtain their seeds of high nutritional value to humans because they contain a high protein percentage rich in the amino acid Lysine. It is also used as green fodder to feed animals. Mung bean is a crop with a comprehensive environmental range and being used as a green fertilizer for its high ability to increase soil fertility and improve soil properties [2]. Most Iraqi soils suffer from a decrease in the availability of many essential soil elements, especially soils located in arid and semi-arid regions. Striving to increase the availability of nutrients to increase the amount of yield is essential, but fertilizing with chemical fertilizers led to the emergence of several problems, the most important of which is environmental pollution. Therefore, the use of biological inoculum and organic fertilizers is currently in a significant increase[3], as they are considered a suitable alternative to chemical fertilizers [4].

Bio inoculation plays a vital role in increasing nutrient availability. They act as a biofertilizer such as the nitrogen-fixing bacteria Rhizobium and as phytostimulators, including microorganisms producing plant hormones such as Azospirillum [5]. They are used as biological control agents such as Trichoderma, which act as defenses to protect plants from plant pathogens [6,7]. The use of biological inoculums reduces the increasing environmental pollution in the soil and the environment in general and their use's economic viability. Rhizobium Leguminosarum is a genus of bacteria that leads to the Chrysogenic family's symbiotic life sympatric to the Leguminous family [8,9]. These bacteria fix atmospheric nitrogen and supply it to the plant in exchange for carbohydrates as a survival and activity source. It equips the plant with nitrogen, phosphorous, and potassium. Trichoderma harzianum is a widespread soil fungus. This fungus is widely exploited in biological control due to the multiplicity of its control mechanisms, including fungal parasitism, antibiotics, enzyme production, competition, and promoting root growth and plant development by increasing the availability of nutrients in the soil [10]. Trichoderma fungi increase plant tolerance to stress by promoting root growth, forming an extensive root system, and tolerating unfavorable environmental conditions. It is considered one of the fungi that enhance plant growth through its role in the cycles of elements, including nitrogen, phosphorous, and sulfur;
in addition to that, it plays a vital role in the solubility of trace elements such as iron, manganese, zinc, and copper in necessary conditions [11,12]. Organic fertilizers have an effective and direct role in increasing soil fertility and productivity and have an essential role in improving soil properties. It prepares the plant with some crucial nutrients that the plant needs during its life. Organic fertilizers should be decomposed entirely, heat-treated, and free of pathogens and weed seeds [13]. Sheep waste has long been proven essential in fertilizing fields. Sheep manure is more critical to crops than cattle manure since sheep manure contains less water and higher organic matter. It is a fast-fermenting and decomposing fertilizer, which significantly improves the soil and reduces its alkalinity [14]. Based on the above, the study aimed to:

1- Study the effect of \textit{R. Leguminosarum} on the number and weight of root nodes and the mung bean crop yield.

2- Study the effect of \textit{T. harzinum} on the number and weight of root nodes and the mung bean crop yield.

3- Study the effect of organic fertilizers (sheep manure) on the number and weight of root nodes and the mung bean crop yield.

4- Study the effect of interaction between \textit{R. leguminosarum} bacteria, \textit{T. harzinum} fungal, and organic fertilizer on the number and weight of root nodes and mung bean yield.

2. Methods and materials

A field experiment was conducted in loamy, sandy soils to grow the crop of mung bean \textit{Vigna radiate} L. during the fall agricultural season 2020 in one of the farms located in Al-Diwaniyah Governorate / Al-Daghara county. A compound soil sample was taken from the field with a depth of (0-30 cm) and mixed well to form a representative soil. The soil was air-dried, then sieved and sifted through a sieve with a diameter of (2) mm, in which some physical and chemical properties were estimated, as shown in Table (1). Seeds and the inoculum: Seeds of the plan, \textit{Vigna radiate} L, a local variety (Wilczek). The Rhizobium (\textit{R. leguminosarum}) inoculum and the Trichoderma (\textit{T. harzinum}) inoculum were used.

2.1. Experimental design and distribution of treatment

A two-factor experiment was conducted according to a Randomized Complete Block Design (R.C.B.D) with three replications, and the experiment included the following treatments:

Factor 1: bacterial inoculum, the application of (B1) bacteria, and a comparison treatment without bacteria inoculation (B0).

Factor 2: fungal inoculum: the application of fungal inoculum (F1) and the comparison treatment without applying the fungus (F0).

Factor 3: organic fertilizer: applying two organic fertilizer levels (5 and 10) t ha\(^{-1}\), a comparison treatment (0).

2.2. Statistical analysis

The data were analyzed statistically for all the studied traits according to the proposed and design used in the experiment using the Gnestat computing program. The L.S.D. The test was used to compare the arithmetic means with a probability of 0.05 (Steel and -ppTorrie, 1980). The field was divided into three main replicates, with (36) experimental units, each with12 experimental sub-units. The experimental unit area is (9) m, and its dimensions are (3 x 3) m. 2m distance was left between the sectors (replications), and (2)m distance was left between the experimental units. The experimental unit included (6) lines, the distance between one line and another is (0.5) m, and the distance between plants within a line is ( 0.5) m.

2.3. Seed inoculation, cultivation, and fertilization

With an area of (806) m\(^2\), the field's land was prepared for plowing, smoothing, and leveling. Organic compost (sheep manure) was applied before planting. The seeds of a local variety (khedrawi) mung bean crop, inoculated with Rhizopian root ganglia and Trichoderma fungi, were planted in the field on the 29\textsuperscript{th} of July 2020 by placing three to five seeds in one seedbed. A week after emergence, the plants were reduced to one plant in the seedbed. Manual weeding was performed to get rid of bushes as needed.
2.4. The studied traits

Five plants were taken randomly from the midlines and for each experimental unit. The following growth and biological characteristics were measured:

1- Number of root nodes of a plant: The roots were extracted from the five samples taken at the flowering stage and randomly selected. They were sieved and washed with a continuous stream of calm water to maintain the knots. The average number of root nodes per plant was calculated [15].

2- Dry weight of root nodes Plant⁻¹: The root nodes are taken from the five plants on which the number of nodes was calculated, were collected and placed in paper bags with the information tagged. They were placed in the electrophoresis at a temperature of (65)°C and for (48) hours. They were weighed using a sensitive balance to extract the plant's dry weight of the root nodules.

3- Dry weight root total: The dry root weights were calculated for samples taken in the stage of full maturity after cutting the shoot of samples. These plants' root system was uprooted, the soil around the roots removed, and then washed under a steady, gentle water stream. They were placed in paper bags and dried in an electric oven at (70) degrees for (48) hours. The dry root weights were calculated and extracted from the root group's average dry weight per plant.

4- Chlorophyll content: Measurement of chlorophyll content in leaves at the flowering stage with Chlorophyll meter-502.

5- Grains yield: It was calculated by taking the rest of the plants' yield in the experimental unit and adding them to the five plants' yield to study the previous traits and obtain the result based on the unit area.

Table 1. Physical and chemical properties of the soil.

| Trait          | Value | Unit          |
|---------------|-------|---------------|
| PH            | 8.09  | --------------|
| EC            | 2.8   | DesiSmens M⁻¹ |
| CEC           | 12.88 | Cml charge kg⁻¹ soil |
| Soil texture  | Sandy Loam |
| Sand          | 556.5 |               |
| Clay          | 53.3  |               |
| Silt          | 390.2 | G kg⁻¹Soil    |
| Organic matter O.M | 0.7   |               |
| N             | 38.22 | Mg kg⁻¹ Soil |
| P             | 2.09  |                |
| K             | 257.82|                |
| Ca            | 14.4  |                |
| mg            | 12.03 |                |
| CO₂           | 0     | Cmol L⁻¹       |
| HCO₃          | 4.75  |                |
| SO₄           | 19.04 |                |

3. Results and discussion

3.1. Number of root nodes (node.Plant⁻¹)

Table (2) presents that inoculation with R.legomnsorium bacteria positively affects the number of root nodes. The treatment of inoculation with Rhizobia (B₁) significantly increased the number of root nodes by (61.17) node Plant⁻¹, while the comparison treatment (B₀) values (36.56) node Plant⁻¹. Because of the inoculation with the active rhizobia bacteria and an increase in their number of nodes in the soil, this is concluded [16]. There was a significant effect of T. harzinum inoculation. The treatment inoculated with Trichoderma (F₁) increased the number of root nodes to (53.00) node Plant⁻¹, while the comparison treatment (F₀) had (44.72) node Plant⁻¹. It is the fungus' role in processing nutrients and the Trichoderma fungus's biological control to resist pathogens [17]. There was a significant increase in the number of root nodes when fertilizing with different levels of organic manure (O₁) (sheep manure), and the highest rate of root nodes was at the level of (5) t ha⁻¹ (56.52) node Plant⁻¹ compared to the comparison treatment (O₀) of (37.83) node Plant⁻¹. It is due to the role of organic matter in the processing of various nutrients,
including nitrogen. It plays a vital role in increasing the number of root nodes. This experiment's results agreed with what was reported [18].

As for the bilateral overlap, the treatment (bacterial inoculation and 5 tons of organic fertilizer) resulted in the highest average in the number of nodes (69.33) node Plant\(^{-1}\), while the comparison treatment, had the least average of (27.00) node Plant\(^{-1}\). It because of the joint role of organic fertilizer and rhizobia bacteria. Organic compost provides the nutrients and organic matter needed by microorganisms and increases organic matter's soil content, increasing the Rhizobia bacteria's activity [19].

The triple interaction (O\(_1\) + F\(_1\) + B\(_1\)) resulted in the highest node numbers' values. It resulted in the highest average in the number of nodes, which (72.33) node Plant\(^{-1}\), while the comparison treatment (O\(_0\) + F\(_0\) + B\(_0\)) gave the least average (24.33) node Plant\(^{-1}\).

### Table 2. The effect of the applied bacteria, fungi, and compost on the number of root nodes (node Plant\(^{-1}\)) at the flowering stage.

| Bacteria (B) | Fungi (F) | Organic matter levels (O) t.ha\(^{-1}\) | Mean of Binary overlap F x B |
|--------------|-----------|------------------------------------|-----------------------------|
|              |           | O\(_0\) | O\(_1\) | O\(_2\) |                     |
| B\(_0\)      | F\(_0\)   | 24.33   | 33.67   | 42.67   | 33.56               |
|              | F\(_1\)   | 29.67   | 52.67   | 36.33   | 39.56               |
| B\(_1\)      | F\(_0\)   | 37.33   | 66.33   | 64.00   | 55.89               |
|              | F\(_1\)   | 60.00   | 72.33   | 67.00   | 66.44               |
| LSD .05      |           | 3.32    |         | 1.92    |                     |
| Bacteria (B) |           |         |         |         | (B) Mean            |
|              | Organic matter levels (O) t.ha\(^{-1}\) |                     |
| B\(_0\)      |             | O\(_0\) | O\(_1\) | O\(_2\) | 36.56               |
| B\(_1\)      |             | 48.67   | 69.33   | 65.50   | 61.17               |
| LSD .05      |             | 2.35    |         | 1.36    |                     |
| Fungi (F)    |             |         |         |         | (B) Mean            |
|              | Organic matter levels (O) t.ha\(^{-1}\) |                     |
| F\(_0\)      |             | O\(_0\) | O\(_1\) | O\(_2\) | 44.72               |
| F\(_1\)      |             | 44.83   | 62.50   | 51.67   | 53.00               |
| LSD .05      |             | 2.35    |         | 1.36    |                     |
| I mean       |             | 37.83   | 56.25   | 52.40   |                     |
| LSD .05      |             | 1.66    |         |         |                     |

### Table 3. The dry weight of root nodes (gm Plant\(^{-1}\))

Table (3) presents that the treatment inoculated with Rhizobia (B\(_1\)) resulted in the highest average weight of the root nodes (0.376) gm Plant\(^{-1}\) compared to the comparison treatment, which gave the least average of (0.299) gm Plant\(^{-1}\). It is attributed to the fact that inoculation with Rhizobia leads to an increase in their numbers in the soil, which can infect the roots and form nodes. It is what was indicated by (Noni, 2012) on the pea plant. There was a significant increase in the root nodes' weight for the inoculation with Trichoderma (F\(_1\)) (0.362) gm Plant\(^{-1}\), while the comparison treatment (F\(_0\)) had with (0.313) gm Plant\(^{-1}\). It is attributed to the fungus Trichoderma's role in decomposing soil organic matter and increasing nutrients' availability [20,21]. The results of Table (3) indicated that the application of organic fertilizer at different levels resulted in a significant increase in the weight of the root nodes, as the highest average weights of the root nodes in the two treatments (O\(_1\) and O\(_2\)) were (0.384 and 0.377) gm Plant\(^{-1}\), Respectively, compared to the comparison treatment (O\(_0\)) (0.252) gm Plant\(^{-1}\). It is attributed to the role of organic fertilizers applied to the soil in increasing the soil's content of organic matter, which leads to an increase in the availability of the nutrients in the soil and consequently to an increase in the weight of the root nodes [22].

The bilateral interaction between biological inoculum and organic fertilizer resulted in a significant increase in root node weight. The treatments of (O\(_1\) + B\(_1\) and O\(_2\) + B\(_1\) yielded the highest averages of root node weight (0.422 and 0.423) gm Plant\(^{-1}\), while the comparison treatment (O\(_0\) + B\(_0\) had the least mean of (0.222) gm Plant\(^{-1}\). It is attributed to the collaborative role between the bacterial inoculum and the compost. The compost increases the organic matter in the soil, which contains essential nutrients for microorganisms. The availability of nutrients and organic matter that organisms need makes microorganisms participate in plant growth with several positive interventions [23]. The triple overlap of the two treatments (O\(_1\) + F\(_1\) + B\(_1\)) (bacterial inoculum +
fungal inoculum + 5 tons of organic fertilizer ha) and \( (O_2 + F_1 + B_1) \) (bacterial inoculum + fungal inoculum + 10 tons of organic fertilizer) resulted in the highest average dry weight of root nodes (0.463 and 0.467 gm Plant\(^{-1}\)), respectively, while the comparison treatment \( (O_0 + F_0 + B_0) \) had the least average (0.207) gm Plant\(^{-1}\).

### Table 3. The effect of the application of bacteria, fungi, and compost on the root nodules' dry weight (gm Plant\(^{-1}\)) at the flowering stage.

| Bacteria (B) | Fungi (F) | Organic matter levels (O) t ha\(^{-1}\) | Mean of Binary overlap F x B |
|--------------|-----------|----------------------------------------|----------------------------|
| B\(_0\)     | F\(_0\)   | 0.207                                  | 0.327                      | 0.313                      | 0.282                      |
| B\(_1\)     | F\(_0\)   | 0.236                                  | 0.367                      | 0.347                      | 0.317                      |
|              | F\(_1\)   | 0.270                                  | 0.380                      | 0.380                      | 0.343                      |
|              |           | 0.293                                  | 0.465                      | 0.467                      | 0.408                      |
| LSD .05     |           |                                        |                            | 0.024                      | 0.014                      |

| Bacteria (B) | Organic matter levels (O) t ha\(^{-1}\) | (B) Mean |
|--------------|----------------------------------------|---------|
| B\(_0\)     | 0.222                                  | 0.347   |
| B\(_1\)     | 0.282                                  | 0.422   |
| LSD .05     |                                        | 0.017   |

| Fungi (F) | Organic matter levels (O) t ha\(^{-1}\) | (B) Mean |
|-----------|----------------------------------------|---------|
| F\(_0\)   | 0.238                                  | 0.353   |
| F\(_1\)   | 0.265                                  | 0.415   |
| LSD .05   |                                        | 0.017   |
| I mean    |                                        | 0.252   |
| LSD .05   |                                        | 0.012   |

3.3. The dry weight of the root mass (gm Plant\(^{-1}\))

Table (4) presents that inoculation with \textit{R. leguminosarum} bacteria has a positive role in increasing the dry weight of the root starch of the plant. Inoculated treatment (B\(_1\)) resulted in a higher average of (1.296) gm Plant\(^{-1}\) compared with a comparison treatment (B\(_0\)) of (0.998) gm Plant\(^{-1}\). The root system increases because the rhizobia bacteria produce many metabolites such as Riboflavin and Cytokine and plant hormones such as gibberellin that elongate and grow the root cells agree with [24], on the pea plant. Table (4) also presented that inoculation with \textit{T. harzianum} significantly affected the roots' dry weight. The treatment inoculated with Trichoderma (F\(_1\)) resulted in an average of (1.242) gm Plant\(^{-1}\), while the comparison treatment, had the least average of (1.053) gm Plant\(^{-1}\). Due to the fungus' role in improving soil construction and increasing the readiness of nutrients and the secretion of a growth-regulating substance and antifungal agents that protect the plant from pathogens, which stimulates the growth of plant roots and increases its weight [25].

There were significant differences in the root mass's dry weight trait due to applying different organic fertilizer levels. The highest value in the treatment was (1.310) gm Plant\(^{-1}\) at the level of 5 tons. Ha, while the least was when the comparison treatment was (0.849) gm Plant\(^{-1}\). The reason is attributed to the importance of the organic matter in improving the physical and chemical properties of the soil, which leads to improving root growth and spread in addition to its role in releasing elements and some active substances for growth, which leads to the formation of a substantial root mass [26].

The two-way interaction, treatment \( (O_1 + B_1) \), resulted in the root starch's highest average dry weight (1.502) gm Plant\(^{-1}\), while the comparison treatment \( (O_0 + B_0) \) resulted in the least average (0.677) gm Plant\(^{-1}\) od root mass. The reason for this is attributed to the joint action between organic fertilizers and rhizobia bacteria. Organic fertilizers work to prepare the plant with the macro and micronutrients it needs and improve the soil's physical and chemical properties, which helps increase root penetration. Rhizobia bacteria decompose organic matter and convert elements into available-to-plant forms, as it promotes plant growth, increases enzymatic activity, and promotes root growth.
As for the triple overlap, treatment (O₁ + F₁ + B₁) (bacterial inoculum + fungal inoculum + 5 tons. Ha of organic fertilizer) resulted in the highest average dry weight of root total (1.547) gm Plant⁻¹, while the comparison treatment resulted in (O₀ + F₀ + B₀) least average (0.553) gm Plant⁻¹ of root mass.

Table 4. The effect of applied bacteria, fungi, and organic manure on the root mass's dry weight (gm Plant⁻¹) in the flowering stage.

| Bacteria (B) | Fungi (F) | Organic matter levels (O) t ha⁻¹ | Mean of Binary overlap F x B |
|--------------|-----------|---------------------------------|-----------------------------|
| B₀           | F₀        | O₀                              | 0.553                       |
|              | F₁        | O₁                              | 0.800                       |
|              | F₀        | O₂                              | 0.907                       |
| B₁           | F₁        | O₁                              | 1.137                       |
|              |           | O₂                              | 1.457                       |
|              |           | Mean                            | 1.250                       |
|              |           | LSD .05                         | 0.023                       |

As for the bilateral interaction, the treatment (O₁ + B₁) resulted in the highest average chlorophyll content in leaves (57.18) SPAD, while the comparison treatment (O₀ + B₀) had the least average of (38.71) SPAD. The increased chlorophyll content in the leaves resulting from the application of organic fertilizer and Rhizobia bacteria inoculation is the joint action in activating many enzymes, including those responsible for building chlorophyll. Also, the rhizobia bacteria perform an important work through their presence in the soil containing organic matter, as it works on decomposing the organic matter into more explicit materials that plants can benefit from [29].
As for the triple overlap, treatment \((O_1 + F_1 + B_1)\) resulted in the highest average chlorophyll content in leaves (59.14) SPAD, while the comparison treatment had the least value (38.19 and 39.22) SPAD, respectively.

| Table 5. The effect of adding bacteria, fungi, and compost on the chlorophyll content in leaves (SPAD) in the flowering stage. |
|---------------------------------------------------------------|
| **Bacteria (B)** | **Fungi (F)** | **Organic matter levels (O) t ha\(^{-1}\)** | **Mean of Binary overlap F x B** |
|------------------|---------------|-----------------------------------|-----------------|
| \(B_0\)         | \(F_0\)       | 38.19 | 43.46 | 46.35 | 42.67 |
| \(B_0\)         | \(F_1\)       | 39.22 | 52.72 | 46.87 | 46.27 |
| \(B_1\)         | \(F_0\)       | 43.73 | 55.22 | 49.26 | 49.41 |
| \(B_1\)         | \(F_1\)       | 46.54 | 59.14 | 56.37 | 54.02 |
| LSD .05         |               | 1.09  | 0.63  |       |       |

| **Bacteria (B)** | **Organic matter levels (O) t ha\(^{-1}\)** | **Mean of Binary overlap O x B** |
|------------------|-----------------------------------------------|---------------------------------|
| \(B_0\)         | \(O_0\) | \(O_1\) | \(O_2\) | (B) Mean |
| \(B_0\)         | 38.71 | 48.09 | 46.61 | 44.47 |
| \(B_1\)         | 45.14 | 57.18 | 52.82 | 51.71 |
| LSD .05         | 0.77  |       |       | 0.44  |

| **Fungi (F)** | **Organic matter levels (O) t ha\(^{-1}\)** | **Mean of Binary overlap O x F** |
|---------------|-----------------------------------------------|---------------------------------|
| \(F_0\)       | \(O_0\) | \(O_1\) | \(O_2\) | (B) Mean |
| \(F_0\)       | 40.96 | 49.34 | 47.80 | 46.04 |
| \(F_1\)       | 42.88 | 55.93 | 51.62 | 50.15 |
| LSD .05       | 0.77  |       |       | 0.44  |
| I mean        | 41.92 | 52.64 | 49.71 |       |
| LSD .05       | 0.54  |       |       |       |

### 3.5. Grain yield (tonnes ha\(^{-1}\))

Table (6) shows that inoculation with rhizobia bacteria resulted in a significant effect in increasing the yield of mung beans. The treatment of inoculation with Rhizobia \((B_1)\) resulted in a higher grain yield rate \((2.821)\ t\ ha^{-1}\) compared to the comparison treatment \((2.050)\ t\ ha^{-1}\). The increase in yield is attributed to the role of rhizobia in forming root nodes, increasing nitrogen preparation, and encouraging root growth, which leads to an increase in the absorption of essential nutrients for the plant, which leads to an increase in nitrogen concentration in the form of protein and amino acids in the grains [30]. There was a significant increase in grain yield in the treatment of inoculation with Trichoderma. The \((F_1)\) inoculation treatment resulted in an average grain yield \((2.582)\ t\ ha^{-1}\), while the comparison treatment had the least grain yield value \((2.290)\ t\ ha^{-1}\). The increase in the growth of the pollinated plants' root system is attributed to the fungus' role in producing growth-stimulating materials and increasing nutrient availability [31]. There were significant differences in the yield of mung beans with the application levels of organic fertilizer. The level of \((5)\ t\ ha^{-1}\) resulted in the highest grain yield \((2.798)\ t\ ha^{-1}\), while the least was for the comparison treatment \((1.862)\ t\ ha^{-1}\). The increase in yield is due to the role of compost in increasing the decomposing organic matter in the soil and its role in improving the physical and chemical properties and fertility and increasing the concentrations of some nutrients, the most important of which are N, P, and K [32]. As for the bilateral interaction, treatment \((O_1 + B_1)\) resulted in the highest average grain yield \((3.314)\ t\ ha^{-1}\), while the comparison treatment \((O_1 + B_0)\) had the least average of \((1.641)\ t\ ha^{-1}\). Organic fertilizer increases the availability of nutrients and increases microorganisms' activity in the soil that decompose organic matter and increase nutrient availability and growth regulators. The increase in yield is a natural result of the overall strength of the root and vegetative mass. The triple interference treatment \((O_1 + F_1 + B_1)\) resulted in the highest average grain yield of \((3.513)\ t\ ha^{-1}\), while the comparison treatment \((O_0 + F_0 + B_0)\) had the least average of \((1.513)\ t\ ha^{-1}\).
Table 6. The effect of the applied bacteria and fungi inoculation and organic manure on the grain yield t ha⁻¹.

| Bacteria (B) | Fungi (F) | Organic matter levels (O) t ha⁻¹ | Mean of Binary overlap F x B |
|--------------|-----------|----------------------------------|-----------------------------|
| B₀           | F₀        | 1.513                            | 2.343                       |
|              | F₁        | 1.770                            | 2.111                       | 2.163                       |
| B₁           | F₀        | 2.001                            | 2.826                       | 2.641                       |
|              | F₁        | 2.163                            | 3.311                       | 3.002                       |
| LSD .05      |           | 0.114                            | 0.066                       |

| Bacteria (B) | Organic matter levels (O) t ha⁻¹ | (B) Mean |
|--------------|----------------------------------|----------|
| B₀           | 1.641                            | 2.282    |
|              | 2.227                            | 2.050    |
| B₁           | 2.082                            | 3.134    |
|              | 3.069                            | 2.821    |
| LSD .05      | 0.081                            | 0.047    |

| Fungi (F) | Organic matter levels (O) t ha⁻¹ | (B) Mean |
|-----------|----------------------------------|----------|
| F₀        | 1.757                            | 2.527    |
|           | 2.584                            | 2.290    |
| F₁        | 1.967                            | 3.069    |
|           | 2.711                            | 2.582    |
| LSD .05   | 0.081                            | 0.047    |
| I mean    | 1.862                            | 2.798    |
| LSD .05   | 0.54                             |          |

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