The Effect of Efficacy Local Isolates of Paenibacillus Bacteria on Available Soil Phosphorus and Growth of Cultivars Wheat (Triticum Aestivum L.)

Ghanim Bahlol Noni and Ayat Hassan Abd

1,2 College of Agriculture, University of Al-Muthanna, Iraq.

Email: bahghanem14@gmail.com

Abstract

A field experiment was carried out to study the effect of bio-inoculation with three local isolates of P. polymyxa and three Varieties of wheat on the growth and yield of the wheat plant (Triticum aestivum L.). In the second agricultural research station affiliated to the College of Agriculture - University of Al-Muthanna, a field experiment was conducted in Al-Muthanna Governorate for the year 2020 in silty clay Loam soil texture. The experiment was designed in a randomized complete block method (RCBD) and with three replications. The experiment included two factors, where the first factor represents the local isolates of four levels, symbolized by P0-P1-P2-P3, and the second factor represents the three wheat varieties, Buhouth class 22, Iraq class, and Ibba 99, which symbolized C1-C2-C3 respectively. The results showed the following: First: The P3 bio-pollination treatment achieved a significant increase in soil phosphorus, potassium, and nitrogen in the available part at the flowering stage, as the highest average was recorded for them (11.39 mg kg⁻¹ soil -1.4300%-1.4.000%), and the P2 treatment was superior for both soil potassium and plant height. And the dry weight, the highest average for them was recorded (157.8 mg kg⁻¹ -92.6 cm - 10.61 g plant⁻¹). Second: As for the cultivars, they differed in most of the characteristics of the study. The Iraq cultivar excelled in soil phosphorus concentration with an average of (11.06 mg kg⁻¹ soil), while Buhouth cultivar 22 was superior in soil potassium with an average of (158.02) mg kg⁻¹ soil. Ibba 99 had been recorded concentration nitrogen and potassium in plants with averages of (1.4208,1.3474%), respectively, while Al-Khrid isolate with Ibba 99 cultivar in nitrogen and potassium concentration plant, where it reached (1.4567-1.4400)/%, while the isolate without addition outperformed with the cultivar Ibba 99 for dry weight, as it reached its highest average (11.40 gm plant⁻¹). The isolate of vegetables also outperformed Iraq cultivar for phosphorous concentration in soil, as it recorded the highest average of (12.30) mg kg⁻¹ soil.

Keywords: Bacteria, Triticum, Growth.

1.Introduction

Biofertilizers are defined as all bacterial inoculant applied to the soil or seeds in many ways with the aim of benefiting from them in preparing some of the necessary nutrients for the plant to improve its growth and increase its productivity. Water and air. Therefore, the world is moving towards clean agricultural technologies with the reduction of pollution as much as possible through the use of natural materials to increase production, such as bio-fertilizers, which are complementary to chemical fertilizers [1]. Organic and biological fertilizers also affect the availability of nutrients, which is one of the important factors in reducing the rate of environmental degradation that results from the continuous use of mineral fertilizers. It also works to improve the quality of the crop at the same time [2], [3], also indicated that use of Bacillus bacteria as a biological inoculum with organic fertilizer leads to an increase in the amount of available-made phosphorous in the soil through the process of improving the chemical and physical properties and increasing the biological activity with a slight decrease in soil PH and bulk density with an increase in the amount of available water and macronutrients. The sum of these effects increases the yield in quantity and quality. Compare with no addition.

The revitalization of the soil microscopic in the rhizosphere plays an important role in promoting plant growth. Recently, interest in studying these organisms has increased. One of the important microorganisms in this field is Paenibacillus...
polymyxa, which was previously known as Bacillus polymyxa, as it excreted the microorganisms that stimulate plant growth and which are found in a high density in the rhizosphere [4]. These bacteria are characterized by a wide range of positive characteristics that promote plant growth, including their ability to fix atmospheric nitrogen as well as their ability to increase the solubility and availability phosphorous in the soil and transform unavailability forms into dissolved form as well as their positive role in improving the physical properties of soil, including improving porosity [5]. Wheat (Triticum aestivum L.) is one of the most important cereal crops grown in the world. Its grains are a basic source of energy needed by humans and are directly included in their diet due to their high nutritional value, as they contain a high percentage of protein and carbohydrates. The results of many studies have shown that it is possible to increase production Wheat crop by adding mineral fertilizers, which provides the needs of this crop through balanced fertilizer combinations [6].

Biomass of bacteria and fungi is critical to improving soil physical and chemical properties and nutrient availability [7]. The introduction of new varieties to the region aims to increase the yield of the unit area, and this matter depends on the appropriateness of the environmental conditions for these varieties, which vary in the degree of their response from one type to another. It leaves a significant impact on the length or shortness of any stage of growth and development [8].

Through the study of the paenibacillus bacteria of the wheat plant, the experiment aimed to study the following:

- Studying the effect of local isolates of P.polymyxa bacteria that stimulate plant growth and their ability to dissolve and prepare phosphorous and convert them from un available forms to dissolved forms.
- Studying the effect of different varieties of wheat crop on the growth and yield of the plant.
- Study the effect of the interaction between local isolates and different varieties on the yield and growth of plants.

2. Materials and Methods

2.1 Location of the experiment

A Factorial experiment was carried out in Muthanna Governorate at the second research station of the College of Agriculture during the winter season of 2020-2021. The soil of the field was of Clay Silty Loam to study the effect of three isolates of P. polymyxa bacteria and three cultivars of wheat (Triticum aestivum) in a randomized complete block design (RCBD). Samples of field soil were taken randomly from three areas of Muthanna Governorate to isolate P. polymyxa bacteria, as they were mixed with each other at each site separately to give a more representative composite sample of the field soil. Then, it was sifted with a sieve whose holes diameter (2 mm) for the purpose of conducting the isolation process, which is related to biological analyzes. Part of the sample may be kept in the refrigerator until assessment.

| Measured characteristic | Unit of measure | Value |
|-------------------------|----------------|-------|
| Ece                     | dsm⁻¹          | 4     |
| pH                      |                | 7.5   |
| Available Nitrogen      |                | 15    |
| Available phosphorous   | mg.kg⁻¹ Soil   | 6.9   |
| Available Potassium     | g kg⁻¹ soil    | 124   |
| Organic matter          | g kg⁻¹ soil    | 1.16  |
| Sand                    |                | 19.5  |
| Silt                    | g kg⁻¹ soil    | 44.3  |
| Clay                    |                | 36.2  |
| Tissue type             |                | silty clay loam |
| Total Bacterial Density | Cfu g⁻¹ soil   | 15*10⁷ |
| P. Polymyxa             |                | 0.14*10¹ |

2.2 Transactions and experimental design

A two-factor experiment was carried out in the field with a randomized complete block design (RCBD) using two factors for the experiment and with three replications distributed randomly. The experiment included the following transactions:

The first factor: the addition of four local isolates of P. Polymyxa bacteria, symbolized by P
• P0 Control
• P1 Applying the isolation of Al-Rumaitha
• P2 Applying the isolation of Samawa
• P3 Applying the isolation of AL-Khudhir district

The second factor: Three varieties of wheat, symbolized by the symbol C

• Bohuth wheat variety 22
• Iraqi wheat variety
• wheat variety Ibaa99

Third: NPK mineral fertilizers, NPK mineral fertilizers were applied according to the complete fertilizer recommendation for nitrogen, potassium and 20% phosphorous and the number of treatment units was 36 experimental units (4 * 3 * 3).

2.3 Preparation of the P. polymyxa inoculant and its use as a biofertilizer

A local isolate of Paenibacillus bacteria bearing the local number (3) and diagnosed of the type (P. polymyxa) was chosen for use in field experiments due to its high efficiency in dissolving phosphorous, and for the purpose of preparing a sufficient amount of inoculant for use in field experiments, three conical flasks with a capacity of (1000) ml containing the Pikovsky medium and inoculum from a prepared liquid culture (50 ml) using sterile pipettes and incubated in the vibrating incubator at a temperature of (30) C for a period of (2-3) days. McFarland standard solution This solution was prepared according to [9], and the density of the used inoculum was (810 * 1.5) colony-forming unit.ml-1. Three varieties of wheat seeds were sterilized with ethyl alcohol for 2 minutes and then washed with distilled water. Several times to remove the remaining sterile material and then put it in sterile glass containers to pour the inoculant with a solution of Arabic gum and peatmoss and leave it for a period of time so that the inoculant and peatmoss stick to the seeds and planted in the field.

2.4 Agriculture and crop service operations

The land was prepared after performing intersecting plowing, smoothing and leveling it, and then the land, which was an area of 500 square meters, was divided into three large sectors (Blocks). The units were divided into eight lines, the distance between one line and another was 20 cm, the width of the plot was 2*2 meter, and the distance between each sector was 1.5 meters to prevent overlapping of the experiment factors. The service operations were carried out according to the need of the crop, and weeding was carried out manually to get rid of the harmful bush, and then the plants were harvested from near the surface, and then they were placed in paper bags for the purpose of the necessary analyzes on them. in the harvest stage.

2.5 Soil measurements

2.5.1 Determination of phosphorous

Phosphorous was extracted using 0.5 N NaHCO₃ as mentioned in Olsen's method, then the color was developed with ammonium molybdate and ascorbic acid and it was estimated by spectrophotometer at a wavelength of 882 nm, solid as mentioned [10].

2.5.2 Determination of potassium

The prepared potassium was extracted with ammonium acetate (N1) solution, and then it was measured using a photometer flame, as mentioned [11].

2.5.3 Nitrogen content in vegetative part (%)

The Nitrogen was estimated according to the method [12], using the Microkjeldal apparatus.

2.5.4 Potassium concentration in vegetative part (%)

The plant extract was estimated by Flame photometer [13].
2.5.5 Dry weight of the vegetable g plant

Ten plants were taken from each experimental unit randomly and the plants were weighed with a sensitive scale to know the dry weight of the plant for vegetative complex after the harvest stage.

2.5.6 plant height (cm)

The height was measured for ten plants selected randomly from each experimental unit, starting from the surface of the soil to the bottom [14].

2.6 Statistical analyses

A factorial experiment was carried out according to a completely randomized block design (RCBD) and the data were analyzed statistically according to the method of analysis of variance using the Genstat discovery edition program. The means were compared using the least significant difference (LSD) method at a level of significance of 0.05.

3. Results and Discussion

3.1 Available phosphorous in soil mg kg$^{-1}$

The results of Table (2) indicate that there are significant differences in the amount of available phosphorous in the soil when bio-inoculation with P. Polymyxa The highest value of available phosphorous was recorded at the P3 treatment, where the highest average was 11.39 mg kg$^{-1}$ soil with an increase of 25.16% compared with the measurement treatment that It gave the lowest average phosphorous concentration of 9.1 mg kg$^{-1}$ soil and the reason for the increase is attributed to the bacterial species P. Polymyxa that have the ability to produce substances that encourage plant growth by absorbing phosphorus and the rest of the other elements by the roots. These results may agree with what [15]. Also, the reason for the increase in phosphorous concentration in the soil is the positive role of the bacteria used [16], indicated to the bacteria Polymyxa P. a high ability to dissolve precipitated phosphates that are not available for absorption into their dissolved forms to benefit the plant for its metabolic activities. As for the effect of the inoculant on the cultivars and the interaction between local isolates and cultivars in the phosphorus concentration in the soil is not significant.

|     | C1  | C2  | C3  | Mean  |
|-----|-----|-----|-----|-------|
| P0  | 8.30| 9.83| 9.17| 9.10  |
| P1  | 10.60| 10.63| 10.63| 10.62 |
| P2  | 12.30| 11.50| 10.10| 11.30 |
| P3  | 11.40| 12.30| 10.47| 11.39 |
| Mean| 10.65| 11.06| 10.09|       |
| LSD | 1.417|    | C=NS|       |

3.2 Available potassium in soil mg kg$^{-1}$

The results of Table (3) indicate that inoculation with P. polymyxa isolates led to a significant increase in the rate of potassium available in the soil, as the highest rate was 157.8 mg kg$^{-1}$soil when the treatment P2, which showed a significant difference from the rest of the isolates with an increase of 9.48 %, while it was less. Average rate of 144.1 mg kg$^{-1}$ soil when compared to treatment. The reason for the increase may be attributed to the role of microorganisms that help potassium from loss processes and increase its availability through the secretion of some enzymes and organic acids. Both of them indicate that the use of biofertilizers increases the necessary nutrients NPK. The results of Table (3) showed that there was a significant effect on potassium availability in soil by the effect of the different varieties, as it gave the highest value for it when treatment C1, as the average for it was 158.02 mg kg-1 soil with an increase of 8.15% compared with the C3 treatment, which gave the lowest value of 146.1 mg kg$^{-1}$ soil [14] stated that adding each of the potassium fertilizers at different levels or mixing them with phosphate fertilizers leads to an increase in the height of the wheat plant and the increase was significant. As for the binary interaction in the same table (3) between local isolates and cultivars, there were no significant differences.
**Table 3.** Effect of inoculation with local isolates of *P. Polymyxa*, and wheat cultivars and the interaction between them on potassium concentration mg kg\(^{-1}\) in soil.

|       | C1     | C2     | C3     | Mean   |
|-------|--------|--------|--------|--------|
| P 0   | 138.7  | 149.7  | 144.0  | 144.1  |
| P1    | 161.0  | 161.0  | 148.0  | 156.6  |
| P2    | 166.7  | 161.0  | 145.7  | 157.8  |
| P3    | 165.7  | 158.0  | 146.7  | 156.8  |
| Mean  | 158.02 | 157.4  | 146.1  |        |
| LSD   | P=10.85| C=9.39 | PC=N.S |        |

3.3 Nitrogen content in the vegetative part (%)

The results of the statistical analysis of Table (4) showed that there were significant differences when inoculated with *P. Polymyxa* isolates in the nitrogen concentration in the available part of the wheat plant, as the treatment P2 significantly outperformed the control treatment, as it recorded the highest average of 1.416% with an increase of 7.59% compared to the control treatment. The measurement that recorded the lowest average amounted to 1.316%, and the reason for the increase in the nitrogen concentration in the available part may be attributed to the role of the organisms used as a biofertilizer in the secretion of growth-promoting hormones such as gibberellin, auxin and indole acetic acid. Therefore, the absorption capacity of nutrients by the plant increases, including the element nitrogen, as well as its ability to fix nitrogen and the necessary elements [17], the results in the same table (4) showed significant differences in the plant nitrogen concentration (%), as treatment C3 gave the highest average of 1.420% with an increase of 4.41% compared to treatment C1 which gave the lowest average of 1.36% and the reason for the increase when using *Paenibacillus* for inoculation, wheat and barley had a significant increase in the growth of these crops and an increase in yield and agreed with [18], that inoculation with bacillus bacteria led to an increase in phosphorous uptake from wheat plant, which led to an increase in the yield, it was found by [19,20], that phosphate-dissolving bacteria PSB helped in the biological fixation of nitrogen and increased the weight and number of root nodes and the availability of some nutrients and then led to an increase in yield and some components of soybean.

The results of the interaction between local isolates and cultivars explain the presence of significant differences, it gave the highest value of 1.4567% in the P3C3 treatment compared to the lowest value in the P0C1 treatment, which gave 1.2700%. Wheat plant and attributed its effect to the production of activated auxin (IAA) by these organisms. [21], also indicated that improving the growth of wheat plant through inoculation with types of associative bacteria *Bacillus* spp., *A. chroococcum*, *Azospirillum brasilense* comes in terms of this production Organisms of growth regulators such as auxins, gibberellins, and cytokinin’s, not because of their processing of nutrients through mineralization and nitrogen fixation processes.

**Table 4.** The effect of inoculation with local isolates of *P. polymyxa* and wheat cultivars and the interaction between them on nitrogen concentration in the vegetable part%.

|       | C1     | C2     | C3     | Mean   |
|-------|--------|--------|--------|--------|
| P 0   | 1.2700 | 1.3233 | 1.3567 | 1.3166 |
| P1    | 1.3667 | 1.4033 | 1.4267 | 1.3989 |
| P2    | 1.3900 | 1.4167 | 1.4433 | 1.4166 |
| P3    | 1.4133 | 1.4200 | 1.4567 | 1.4300 |
| Mean  | 1.3600 | 1.3908 | 1.4208 |        |
| LSD   | P=0.0116| C=0.0100| PC=0.0201|        |

3.4 Potassium concentration in vegetative part (%)

The results indicated in Table (5) the statistical analysis of the effect of bio-inoculation with *P. Polymyxa* bacteria on the vegetative part of the wheat plant. The P3 treatment achieved the highest average of 1.400%, with an increase of 18.14% compared to the measurement treatment, which gave the lowest average of 1.185%. This treatment P3 exceeded the rest of the treatments due to the increase in the number of bacteria, which in turn improves the physical properties of the soil compared to its numbers far from the rhizosphere, which leads to improving the soil and providing the nutrients necessary for plant growth, including potassium, in addition to the difference in the mechanism of microorganisms secretion of hormones and enzymes. These results are indicated by [22].
The results also showed in the same table that there were significant differences in the concentration of potassium in vegetative part due to the influence of different varieties, as it gave the highest value to it when treatment C3, the highest average amounted to 1.347%, with an increase of 6.81% compared to treatment C1, which gave the lowest average amounted to 1.261%. The reason for the increase is that inoculation of wheat plants with P. Polymyxa bacteria increased the average diameter of soil particles as a result of this bacteria secreting the polysugar materials, which lead to linking the particles with each other and improving the construction of sandy soil, this was indicated by [23]. It was noted that the interaction between local isolates and cultivars does not have significant differences between them.

Table 5. The effect of inoculation with local isolates of P. polymyxa and wheat cultivars and the interaction between them on potassium in the vegetable part %.

|     | C1   | C2   | C3   | Mean |
|-----|------|------|------|------|
| P 0 | 1.1300 | 1.1933 | 1.2333 | 1.1855 |
| P1  | 1.2567 | 1.2733 | 1.3333 | 1.2877 |
| P2  | 1.3033 | 1.3500 | 1.3833 | 1.3455 |
| P3  | 1.3567 | 1.4033 | 1.4400 | 1.4000 |
| Mean| 1.2616 | 1.3047 | 1.3474 |      |
| LSD | P=0.0190 | C=0.0164 | PC=N.S |

3.5 Plant Height (cm)

The results showed in Table (6) that there were significant differences for the plant height characteristic of wheat plant inoculated with Polymyx P. The treatment P2 excelled with the highest average of 92.6 cm with an increase of 81.41% compared with the measurement treatment that gave the lowest average of 78.2 cm. The reason for the increase is attributed to that the increased biological activity as a result of inoculation with bacteria leads to a decrease in soil pH in the rhizosphere, which facilitates the processes of absorption of elements in general, the smallest property of them, and this reflects positively on improving the characteristics of the plant. Organic acids and phosphatase enzyme, in addition to their secretion of growth-regulating substances such as indole, phenol, giberellin and cytokines, which in turn stimulate plant growth, which led to an increase in this trait [24]. As for the cultivars, there is no significant effect of wheat plant height.

The results in the same table also indicate the bilateral interaction between local isolates and cultivars, indicating that there are significant differences in plant height, as the highest value was recorded at 99.8 cm when treatment P2C1 increased by 48.06% compared with treatment P1C1, which gave the lowest value of 67.2 cm. The additive prepares phosphorous and secretes antibiotics, in addition to its secretion of IAA, which affects cell elongation and division. By these mechanisms, the formation of protein substances increases, which is positively reflected in the rate of plant growth. This result is consistent with what was indicated by [25].

Table 6. Effect of inoculation with local isolates of P. polymyxa and wheat cultivars and the interaction between them on plant height of wheat (cm).

|     | C1   | C2   | C3   | Mean |
|-----|------|------|------|------|
| P 0 | 94.5 | 83.8 | 99.5 | 86.8 |
| P1  | 67.2 | 85.2 | 82.4 | 78.2 |
| P2  | 99.8 | 77.6 | 83.0 | 92.6 |
| P3  | 85.3 | 89.9 | 90.7 | 88.6 |
| Mean| 86.7 | 84.1 | 88.9 |      |
| LSD | P=16.55 | C=N.S | PC=28.67 |

3.6 Dry weight of the vegetable g plant^{-1}

The results of Table (7) showed that there were significant differences in the dry weight characteristic of vegetative group of wheat when using P. Polymyx bacteria as a live inoculum. It gave the highest average when treatment P2 reached 10.61 gm plant^{-1} with a significant difference from isolate P0, which gave it the same average that 10.40 gm plant^{-1} was recorded, with an increase of 10.98% compared to the measurement treatment, which reached the lowest average of 9.56 gm plant^{-1}. The reason for the increase in the dry weight of vegetative mass when the plant was inoculated with P.polymyxa isolates may be due to the role of those organisms in secreting growth hormones such as Giberellins, indole, and organic acids that dissolve and prepare nutrients from their insoluble compounds in the soil, in particular the provision of phosphorous, which supplies
the plant with energy and helps cell division in the plant, and this is reflected positively on the growth of the plant, including the characteristic of the increase in the dry weight of vegetative group in the plant [26,27].

As for the cultivars, there was no significant effect on the dry weight characteristic of vegetative total of the wheat plant. the results also indicate in the same table (7) significant differences for the binary interaction between cultivars and isolates, where the highest value reached 11.40 gm when treatment POC3, with a significant difference from the two isolates P1C1 and P2C1 which recorded 11.00 and 11.03 gm plant$^{-1}$ sequentially with an increase of 38.51% compared to With the lowest value recorded, 8.23g of plant$^{-1}$ and the reason for the increase is due to the action of microorganisms added with different mechanisms and, its including the dissolution of some nutrients from their insoluble compounds in the soil, as well as the secretion of some organic acids, some hormones and growth regulators that affect cell division and stimulate plant growth and support. These secretions include plant growth, including an increase in its dry weight [28]. She agreed with [29], the role of biological fertilization in increasing the availability of nutrients as well as the secretion of growth stimulants. [30], also indicated an increase in dry weight because the pollen coats the seeds, as well as an increase in the concentration of pollen in the rhizosphere surrounding the roots, and then an increase in the chance of root infection. And then the effectiveness of the inoculant decreases and this was supported by [31], in a study he conducted that included the effect of the method of adding bio-fertilizers in increasing the effectiveness of the inoculant.

**Table 7.** The effect of inoculation with local isolates of *P. polymyxa* and wheat cultivars and the interaction between them on the dry weight of vegetative total of wheat plant.$^{1}$

|       | C1   | C2   | C3   | Mean |
|-------|------|------|------|------|
| P 0   | 9.03 | 10.77| 11.40| 10.40|
| P1    | 11.00| 10.47| 8.23 | 9.90 |
| P2    | 11.03| 10.43| 10.37| 10.61|
| P3    | 10.70| 8.37 | 9.60 | 9.56 |
| Mean  | 10.44| 10.01| 9.60 |      |
| LSD   | P=0.875 |     | C=N.S | PC=0.875 |

**References**

[1] Al-Jubouri, Abdullah Karim Jabbar. (2012) . The effect of bio-inoculation with Azotobacter chroococcum bacteria, Glomus mosseae and phosphate rock on phosphorous availability and wheat plant growth, Master’s thesis, College of Agriculture, Al-Muthanna University.

[2] 2. Noni, Ghamim Bahlol. (2016). The effect of the type of bio-fertilizer Paenibacillus polymyxa and Glomus mosseae and the method of its addition and the type of yield on the availability of phosphorus in the soil and the growth and yield of yellow corn, PhD thesis, College of Agriculture, University of Baghdad.

[3] Al-Absawy, Mohamed Hassan Meleh. (2018) . The effect of adding different levels and batches of phosphate fertilizer on the kinetics of phosphorous release in the soil and in the growth and yield of two wheat cultivars. Master’s Thesis, College of Agriculture, University of Al-Muthanna.

[4] Hassan, Zainab Kazem (2011). Isolation and identification of Azospirillum lipoferum and Bacillus Polymyxa from some soils in southern Iraq and their role in biofertilization of maize plants (Zea mays L.). PhD thesis, Faculty of Agriculture, Al-Basra university.

[5] Saad, Turki Muffen and Ghanim Bahlol Noni, Ahmed Hamid Mazyad. (2020) . The effect of bio-inoculation with two local isolates of Paenibacillus Polymyxa bacteria and levels of organic matter on NPK avialability, growth and productivity of maize. Master’s thesis, College of Agriculture, University of Al-Muthanna.

[6] Al-Manama, Abdel Raouf Ali. Ph.D. in Microbiology, Islamic University of Gaza, and Rawan Hassan Reeda, Master’s degree in Microbiology, Islamic University of Gaza, Arab Scientific Society Organization 2017.

[7] Harran, Muhammad Saeed. (2014) . Effect of phosphate-dissolving fungi and organic matter on dissolving phosphate rock and their effect on wheat plant growth, Technical Education Authority.

[8] Al-Shamy, Asmaa Salim Hussein. (2011). The effect of biological, organic and mineral fertilization (Azotobacter) on the growth and yield of bread wheat and its nutrient content, Master's thesis, College of Agriculture, University of Baghdad.

[9] Al-Barakat, Hanoun Nahi Kazem. (2016) . The effect of biofertilization and methods of adding humin and fulvic acids on the avialability of NPK, iron and zinc in the soil and the productivity of maize.

[10] Kumar, Vivek. (2010) . A lecture on biological plant fertilizers, the optimal alternative to chemical fertilizers, Public Authority for Agricultural Affairs, Kuwait. Al-Qabas newspaper, issue 13-464.

[11] Al-Rifai, Shaima Ibrahim Mahmoud. (2006). Response of wheat cultivars to foliar feeding with iron and manganese, PhD thesis, College of Agriculture, University of Basra.

[12] Al-Shamy, Monem Fadel Mosleh. (2007). The effect of biofertilization with Glomus mosseae and Trichoderma harzianum and organic fertilization with humin acid and the interaction between them on plant growth. Master’s thesis. Council of the Higher Academy of Scientific and Human Studies. Department of Life Sciences.
[13] Al-Zoghbi, Muhammad Manhal. (2006). The effect of decomposing microorganisms for phosphate and organic matter on the dissolution of phosphate rock on crop productivity, PhD thesis, Faculty of Agriculture, Damascus University.

[14] Al-Falah, Mahmoud Howaidi. (1988). Effect of levels and methods of adding phosphate fertilizer on available phosphorus and plant growth in desert soils. Master's thesis, College of Agriculture and Forestry, University of Baghdad.

[15] Alguacil, M. M.; E. Caravaca; A. Roldan. (2005). Changes in rhizosphere microbial activity mediated by native or allochthonous AM fungal in the reforestation at

[16] Ash, C., Priest, F.G., Collins, M.D. (1993). Molecular identification of rRNA group 3 bacilli (Ash, Farrow, Wallbanks and Collins) using a PCR probe test. Antonie van Leeuwenhoek 63, 253±260

[17] Fouad Houssein Kamel. "Development of in-vitro Sensitivity Testing for Pathogenic Bacteria". Al-Qadisiyah Journal For Agriculture Sciences, 8, 1, 2018, 1-5. doi: 10.33794/qjas.2018.162643

[18] Olsen, S.R. and L.E. Sommers (1982). Phosphorus in A.L Page, (Ed). Methods of Soil Analysis. Part2. Chemical and Microbiological Properties 2nd edition. Amer. Soc. of Agron. Inc. Soil Sci. Soc. Am. Inc. Madison, Wis. U.S.A.

[19] Page, A.L.; R.H. Miller and D.R. Keeny. (1982). Methods of Soil analysis part (2) 2nd (ed). Agronomy 9. Amer. Soc. Agron. Madison Wisconsin.

[20] Black, C.A. (1965). Methods of soil analysis .Amer.soc. of agron. Inc. USA. 20.

[21] Bremner, J.M. 1965. Inorganic forms of nitrogen in C.A. Black . 1965 . Methods of soil analysis . amer . Soc. Of Agron. Inc. USA.

[22] Sundara, B.; V. Natarayan and K. Hari (2001). Influence of phosphorus solubilizing bacteria on soil available P - status and sugar cane development on a tropical vertisol.

[23] Arti. S. Shanware: Suresha. A. Kalkar. and Minal . M. Trivedi. (2014). Potassium solubilizer : occurrence mechanism and their role as competent biofertilizers. International J. of Current Microbiology and Applied Sciences, ISSN.

[24] Bezzate, S.; S. Aymerich ; R. Chambert ; S. Czarnes ; O. Berge and T. Heulin (2000). Disruption of the Bacillus ploymyxa levansucrase gene impairs its ability to aggregate soil in the wheat rhizosphere, Enviro. Microbial.

[25] Salama T. Al-Musawi; Faisal M. M. Al-Tahir. "The Effect of the Treatment of Foliar Different Nutritional and Hormonal on the Growth Yield of Four Varieties of Vica faba L.". Al-Qadisiyah Journal For Agriculture Sciences, 8, 1, 2018, 27-34. doi: 10.33794/qjas.2018.162650

[26] Son , T.T.N. ; C.N. Diep and T.T.M. Giang. (2006). Effect of bradyrhizobia and phosphate solubilizing bacteria on the germination of cicer arrietinum seeds and seedling growth

[27] Brown , J.W. (2001). Enrichment & isolation of Bacillus

[28] Afzal, A.; A. Ashraf ; A. Saeed and M. Farooq . (2005). Effect of phosphate solubilizing microorganism on phosphorus uptake , yield and yield traits of wheat ( Triticum aestivum L ).

[29] Ponmurugan, P. and C. Gopi (2006). In vitro production of growth2 29.

[30] regulators of phosphatase activity by phosphate solubilizing bacteria

[31] Haynes, R.J. (1980). A comparison of two modified kjeldhal digestion techniques for multi - element plant analysis with conventional wet and dry ashing methods. Communication in Soil Sci. Plant Analysis.