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Effects of Plant Growth-Promoting Rhizobacteria (PGPR) Consortia on Wheat Seed Germination and Growth Enhancement

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***Abstract*— PGPR consortia are Growth promoting group of bacteria that make plant growth better by helping to dissolve phosphates, produce siderophores, and synthesizing Indole Acetic Acid (IAA). The study was designed to check how 4 PGPR Consortia (T1, T2, T3 and T4) with different growth promoting qualities impact the growth of wheat under set conditions. Eleven PGPRs were isolated and characterized from the rhizosphere for important PGPR abilities and also tested strains that were compatible to form consortia. The seeds that had been treated showed higher germination, stronger development of roots and shoots, more chlorophyll, and more growth than the untreated control seeds. The root length of wheat plant treated with consortium T1were highest (27.12 cm) as well as their shoot length (59 cm) and chlorophyll content (5.593 mg/g). wheat plant treated with consortium T1, the shoot had a 99% increase in leaves when compared to the 3.65% in control. The findings show that PGPR consortia, mostly wheat plant treated with consortium T1 and T3, may work well as natural fertilizers to boost the sustainability of wheat farming. Farmers will benefit from more field trials to check if the methods work in different climates..**

***Keywords*— PGPR, Wheat, Rhizobacteria, Seed Germination, Biofertilizer, Bacterial Consortia**

I. INTRODUCTION

Wheat (Triticum aestivum L.) is a muesli crop of global implication, providing about 20% of total dietary calories and protein intake by the human race. With the growing demand because of population pressure, enhancing wheat yield in a sustainable way has emerged as an urgent agenda. Conventional farming methods depend largely on chemical fertilizers, which have the potential to cause soil deterioration and pollution (De Andrade et al. 2023). Thus, eco-friendly alternatives such as PGPR have been under the spotlight since they have the potential to boost the productivity of crops without causing deterioration of the soil.

PGPR consortia are soil bacteria that inhabit ate the root space of a plant and elicit growth by several mechanisms including nitrogen fixing, phosphorus solubilization of insoluble sources, phytohormone production especially Indole Acetic Acid (IAA) and siderophore production that increase iron availability.

In vitro compatibility tests allowed the screening of eleven bacterial isolates and their combination into four consistent consortia (T1 -T4). These formulations were tried both on petriplate and pot conditions.

II. MATERIAL AND METHODS

2.1 Screening and Quantification of PGPR Traits

Out of the eleven isolates, screening was done for phosphate solubilization, siderophore formation, and Indole Acetic Acid (IAA) production in each isolate. To find out how well the strains solubilized phosphate, Pikovskaya's agar was used. Clear halo zones appeared around colonies that were able to solubilize phosphate. In liquid Pikovskaya's broth, quantitative analysis was completed to determine solubilized phosphate with the molybdenum blue method (Chieb and Gachomo, 2023). To check for siderophore activity, yellow-orange zones formed around CAS agar while the darkening of CAS solution measured at 630 nm to quantify. Isolates were cultured in Luria-Bertani broth together with L-tryptophan, and activity of the enzyme was assessed by Salkowski's reagent color; finally, the intensity of pink coloration was checked spectrophotometrically at 530 nm (Khoso et al. 2024). The information from these experiments demonstrated which bacteria offered the most help to plant growth, so those were used to prepare the consortia for further work.

2.2 Compatibility Test Assay

Mutual compatibility between eleven rhizospheric bacteria was checked by using the perpendicular streak method on nutrient agar plates (Hyder et al. 2023).

Every isolate was placed at right angles to the others and each plate was kept at 28°C ± 2°C for 2 days. For each combination, the zones of interaction were marked with "I" if there was an inhibition and with "0" if there was compatibility. Based on the compatibility between specific isolates, T1, T2, T3, and T4 consortium were prepared from selected PGPR for further experiments.

2.3 Retrieval of Bacterial Isolates

First, the samples needed to be revived and grown before using them in seed treatment and soil application. One hundred microliters from the glycerol stock of chosen bacterial isolates were mixed into 50 mL of sterile nutrient broth and then gestated at 28 ± 2°C for 24 hours with shaking, to be sure that the bacteria were well grown and active. With the help of a spectrophotometer and measuring at a wavelength of 600nm, the suspension was adjusted to about 10^8 CFU/mL. For two different reasons, these growing cultures were used: (i) to treat surface-sterilized wheat seeds with the suspension so that all the seeds were coated evenly, and (ii) to apply 2 mL of the culture solution close to the roots of the wheat in the study pots (Hasan et al. 2024). Grown cultures were kept at 4°C for quick use then in glycerin stocks at –20 °C to preserve.

2.4 Seed Germination Assay (on Petri Plate).

Wheat seed germination assay was performed using prepared PGPR consortia on Petri plate in the laboratory. Before planting, healthy wheat seeds were sterilized in a resolution of 0.1% $HgCl_2$ for few minutes and then well wash by sterilized distilled water. All the sterilized seeds were then treated with freshly prepared suspensions of the T1, T2, T3, and T4 consortia, separately so they would be coated evenly for 1 hour. Some seeds were treated with distilled sterile water and used as the 'control' group (Zahra et al. 2023). The seeds that had been treated were put on sterile filter paper inside Petri dishes that were moistened with distilled water and stored at 20°C. It checked the germination every day for six days, and it considered the appearance of the radicle as a sign of germination (Salem et al. 2024). The progress in germination and growth of seed roots and shoots were measured for each consortium.

2.5 Pot Trial

2.5.1 Soil Preparation:

Pot trial was setup to study how different PGPR consortia influenced the growth of wheat in a controlled environment. The soil used in the experiment was picked from an agricultural field and was studied to find out its physicochemical properties before being put into use.

The soil's pH measured as 7.9, which means it is suitable for wheat farming. Among the available nitrogen, phosphorus, and potassium, the field was found to have 215 kg/ha, 110 kg/ha, and 390 kg/ha, which represents it was suitable for cultivation. According to the analysis, the amount of organic carbon present in the soil was 0.75% which was enough for microbial and root growth (Singh et al. 2023). Then the soil was sterilized to remove

Physiochemical Parameter of the soil:

- pH: 7.9
- Available Nitrogen: 215 kg/ha
- Phosphorus: 110 kg/ha
- Potassium: 390 kg/ha
- Organic Carbon: 0.75%

2.5.2 Seed treatment and Potting

To examine how well PGPR consortia work in soil, the experiment was done by applying them directly onto sterile seedling trays in soil (Mdpi.com, 2025). The wheat seeds were sterilized and then planted in Pots full of treated and sifted soil as used in the pot trial. In each case, seeds were treated with one of the PGPR consortia (called T1 to T4) and immediately after sowing, 2 mL of each PGPR suspension was applied close to the roots. Autoclaved Distilled water was used as the control for the seeds (Mahreen et al. 2023). Each treatment was done three times to make the study results statistically reliable. They were kept under standard conditions such that the temperature was 24 ± 2°C and the light intensity was 1000 lux.

2.5.3 Measurement of Morphological and Biochemical Parameters:

Morphological parameter: Fresh weight, Dry Weight, shoot length, root length, No. of Leaves % were measured after 40 days, 80 Days, 120 Days after germination.

Total Chlorophyll, chlorophyll A and Chlorophyll B from the leaf sample were estimated using the DMSO method (Blanke, 1992). A 100 mg fresh plant leaf sample was placed in a 100 ml volumetric flask containing 10 ml DMSO (Dimethyl sulfoxide). The flask was allowed to stand overnight before measuring the spectroscopic absorbance at 663 and 645 nm. DMSO without leaf sample was used as blank, and un-inoculated wheat leaf was used as control. The total chlorophyll content of the wheat leaf was calculated using the formula given below.

2.6 Molecular characterization of the rhizospheric isolates

Genomic DNA was immediately removed since each sample following the standard procedure that involves phenol-chloroform.

A Universal Amplification from 16S rRNA with primers 27F and 1492R was done by PCR, and then sequencing was carried out. BLAST analysis was performed to look for similarities of the sequences in the GenBank database from NCBI.

III. Results And Discussion

3.1 Consortium preparation:

The experiments of eleven rhizospheric bacterial isolates using the cross-streak assay discovered mutually compatible combinations that can be used in forming a consortium. Lack of any inhibition areas between some isolate's points to the possibility that they can cooperate and work observed, four consortia were made to be used in germination and pot trials. Among them, you had: T1 (VS4S1 to VS4S2 to VS6K3), T2 (VS4S1 to VS2S3 to VS3S3), T3 (VS6K3 to VS4S2 to VS2S3), and T4 (VS6A1 to VS4S1 to VS2S3). It designed every consortium with strains that have different plant growth-promoting abilities, such as solubilizing phosphate, producing siderophores, and making IAA (Gaspareto et al. 2023). The choices were made to maximize the nutrients for wheat and make sure no two strains were competitive so that their usefulness for developing a healthy wheat crop increased.

3.2 Seed Germination Assay (on Petri Plate)

Seed Germination Assay on Petri dish test, all seeds treated with four prepared consortia separately showed much better germination than those in the control. Wheat seeds process with consortia T1 to T4 germinated sooner, were more uniform, and were measured as having a higher vigor index. The use of PGPR helped boost the development of the leaves of plants at the beginning of their development. The best effects were seen in consortium T1 (VS4S1-VS4S2-VS6K3), as on the six day after germination its average root length was 4.8 cm and average shoot length was 3.0 cm—higher than in the control, which had only 1.4 cm for roots and 1.6 cm for shoots which is supported by Shankar and Prasad, 2023.

3.3 Pot Trial

3.3.1Measurement of Morphological and Biochemical Parameters:

Control Treatment

Treatment 1

Treatment 2

Treatment 3

Treatment 4

Figure1: Pot Trial

3.3.1.1 Shoot Length (cm)

Wheat treated with PGPR consortia had significantly increased shoot lengths as revealed by the pot study.

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Forty-day after germination, plants treated with T1 consortia had the most shoot growth, at 25 cm, while Wheat plant treated with consortium T3 measured 20 cm and control plants measured 19 cm. Wheat plant treated with consortium T1 progressed well and was significantly longer at 56 cm by 80 days after germination, more than Wheat plant treated with consortium T2 (44 cm), Wheat plant treated with consortium T3 (39 cm), and Wheat plant treated with consortium T4 (34 cm), and compared to 35 cm in the control. On day 120 after germination, Wheat plant treated with consortium T1, length was 59 cm, and both Wheat plant treated with consortium T3 and Wheat plant treated with consortium T4 came close at 53 cm and 54.33 cm, respectively (Masmoudi et al. 2023). These findings prove that beneficial effects of T1 PGPR consortium are noticeable in the shoot growth of the plant during its development in controlled pots.

Table 1:
Shoot Length (cm)

Days	Control	T1	T2	T3	T4
40	19	25	18	20	17.5
80	35	56	44	39	34
120	53.5	59	48	53	54.33

3.3.1.2 Root Length (cm)

When wheat was treated with PGPR consortia, its root growth was much better than in the control sample. Forty days after germination, roots of all consortia had increased in length, and Wheat plant treated with consortium T4 recorded the highest at 5.15 cm, succeeded by Wheat plant treated with consortium T1 and T2 at 5.00 cm, while the control Root length was 4.03 cm. At 80 days, the roots of Wheat plant treated with consortium T2 were the longest, measuring 12.11 cm, and the roots of Wheat plant treated with consortium T4 and T3 were 9.34 cm and 9.12 cm, respectively, while the control group's roots were the same as before at 8.66 cm. Twelve weeks after the experiment started, Wheat plant treated with consortium T1 measure 27.12 cm in root length, beating Wheat plant treated with consortium T2 (23.12 cm), T3 (18.13 cm), and T4 (18.66 cm), both of which were higher than what was measured in the control sample of just 12.19 cm.

3.3.1.3 Root Length (cm)

Table 2:
Root Length (cm)

Days	Control	T1	T2	T3	T4
40	**4.03**	**5.00**	**5.00**	**4.63**	**5.15**
80	**8.66**	**7.12**	**12.11**	**9.12**	**9.34**
120	**12.19**	**27.12**	**23.12**	**18.13**	**18.66**

3.3.1.3 Biomass Production

Fresh Weight (g)

PGPR treatments led to a higher fresh weight of wheat plants in the different stages of growth. Wheat plant treated with consortium T1 after 40 days of germination resulted in the highest recorded fresh weight, with 5.0 gm, which was much higher than the control's 2.64 gm. Although control was at its highest (5.75 gm) after 80 days of germination, Wheat plant treated with consortium T1 kept improving steadily, ending with 5.8 gm after 120 days after germination. Wheat plant treated with consortium T2 and T3 brought about increases in fresh biomass in comparison to the control, with the Wheat plant treated with consortium T1 being highest and fairly stable.

Table 3:
Fresh Weight (gm)

Days	Control	T1	T2	T3	T4
40	2.64	5.0	4.3	3.0	2.9
80	5.75	4.8	4.1	3.1	3.7
120	3.78	5.8	4.8	4.7	3.6

Dry Weight (gm)

Plants treated with PGPR grew much bigger than the control plants. After 40 days of germination, Wheat plant treated with consortium T4 had the biggest dry weight (1.87 gm) while the control had very little, just 0.14 gm. At the end of 120 days after germination, the Wheat plant treated with consortium T4 had its peak at 3.9 gm, and Wheat plant treated with consortium T2 had reached 3.12 gm. According to the results, T4 and T2 consortia helped improve biomass production during the growth of the plants.

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Table 4:
Dry Weight (gm)

Days	Control	T1	T2	T3	T4
40	0.14	1.00	0.19	0.18	1.87
80	1.13	2.11	2.17	2.01	1.98
120	1.68	2.00	3.12	2.00	3.9

3.3.1.4 Number of Leaves (%)

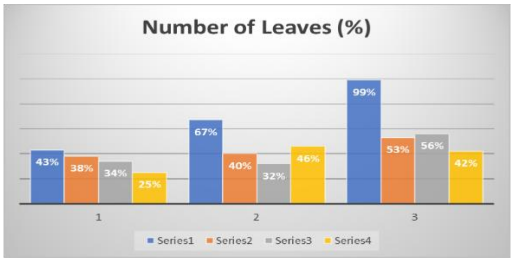

Figure 2: Number of Leaves

When leaves were compared to the control, the percentage increase in wheat plants treated with PGPR consortia was noticeably better. At 40 days after germination, the Wheat plant treated with consortium T1 recorded 43%, which was more than the 5.34% seen in the control. At the end of 80 days after germination, Wheat plant treated with consortium T1 marked the largest growth again (67%), while Wheat plant treated with consortium T4 showed a moderate one (46%). At the 120-days after germination, Wheat plant treated with consortium T1 demonstrated almost perfect leaf production, showing it had the best vegetative growth compared to the rest (Voronina et al. 2023). On the whole, Wheat plant treated with consortium T1 gave the best outcomes, proving that it successfully promoted development of new leaves.

Table 5:
Number of Leaves (%)

Days	Control	T1	T2	T3	T4
40	5.34%	43%	38%	34%	25%
80	6.43%	67%	40%	32%	46%
120	3.65%	99%	53%	56%	42%

3.3.1.5 Chlorophyll Content

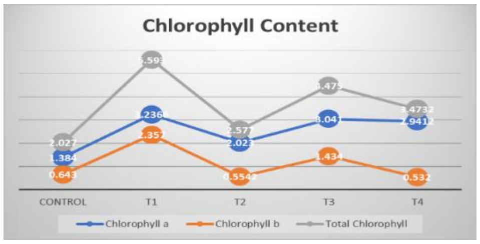

Figure 3: Chlorophyll Content

The application of PGPR consortia changed the wheat leaves' chlorophyll content and made the plants have better photosynthetic activity and more vigor. Wheat plant treated with consortium T1 had the most growth seen in its chlorophyll, with 3.2366 mg/g for chlorophyll a, 2.357 mg/g for chlorophyll b, and 5.593 mg/g in total chlorophyll. Compared to the treatment group, the chlorophyll a for the control was 1.384 mg/g, the chlorophyll b was only 0.643 mg/g, and total chlorophyll came to 2.027 mg/g. Even though Wheat plant treated with consortium T3 and T4 did not match Wheat plant treated with consortium T1's level of chlorophyll content, they still had higher amounts than most other samples (Shahwar et al. 2023). Improved chlorophyll in treated plants is perhaps the result of PGPR contributing phosphorus to plants and producing IAA (Irfan et al. 2025). From the study, it is evident that PGPR support both root and shoot development and boost the traits important for plant health, and T1 consortium is the best choice for raising photosynthetic efficiency in wheat.

Table 6:
Chlorophyll Content (mg/gm)

Treatment	Chlorophyll a	Chlorophyll b	Total Chlorophyll
Control	1.384	0.643	2.027
T1	3.2366	2.357	5.593
T2	2.023	0.5542	2.577
T3	3.041	1.434	4.475
T4	2.9412	0.532	3.4732

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3.4 Discussion

It is obvious from the study that applying PGPR consortia to wheat can promote its growth and healthy development. Seed germination, growth of roots and shoots, chlorophyll amount, and biomass were all found to be higher in the PGPR consortia-treated group than in the control that saw no treatment (Dembitsky et al. 2022). Wheat plant treated with consortium T1 and T3 gave the best and most noticeable results for most of the parameters measured. So, using several bacterial strains together that share many plant growth-promoting features can greatly improve crop productivity.

Wheat plant treated with consortium T1 grew bigger roots in the Petri dish, increasing length from 1.4 cm to 4.8 cm, and taller shoots from 1.6 cm to 3.0 cm. This research proves that using PGPR to treat plants increases IAA phytohormone levels, which speed regeneration of plant cells right after germination (Areej et al. 2024). Wheat plant treated with consortium T1 also had greater chlorophyll, causing total chlorophyll to go from 2.027 mg/g in control to 5.593 mg/g, indicating improved productivity and metabolism.

More information from pot studies adds to the positive effects of PGPR. After 120 days, roots of Wheat plant treated with consortium T1 and T2 measured 27.12 cm and 23.12 cm, far longer than those of the regulator by a length of 12.19 cm. Similarly, the treatment under T1 resulted in 59 cm of shoot length, which is longer than what the control produced at 53.5 cm. These improvements might happen because the bacterial groups can now absorb nutrients more efficiently with help from phosphate solubilization and iron from siderophores.

This information was supported by data from biomass measurements, as well. Around 120 days after the germination, the dry weight in Wheat plant treated with consortium T4 (3.9 g) was more than two times higher than the dry weight in the control (1.68 g). While there was just a 3.65% growth in the size of leaves in the control, the treated plants saw a 99% rise (Lastochkina and Bosacchi, 2023). These results indicate that microbial groups can strengthen growth at the start and boost productivity all through the crop's life.

IV. Conclusion

Using PGPR consortia increased most of the growth parameters in wheat when grown in a controlled environment. Neither chemical nor genetic treatments gave better results than the control when it came to seed germination, chlorophyll content, and how long the roots or shoots grew.

Wheat plant treated with consortium T1 had a root length of 27.12 cm, a shoot length of 59 cm, and a total chlorophyll content of 5.593 mg/g by 120 days, which is much higher than the amount found in the control group. Such progress is because of the interactions between microbial talents like phosphate solubilization, siderophore production, and the synthesis of IAA.

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