

## Enhancing Tef Growth and Yield through Integrated Application of Plant Growth-Promoting Rhizobacteria and Reduced Chemical Fertilizers

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### ABSTRACT

Tef is a staple crop in Ethiopia, serving as the primary food source for approximately 70 million people and contributing significantly to the country's agricultural economy. Its high protein content, gluten-free nature, and rich micronutrient profile make it nutritionally vital for populations with dietary restrictions like celiac disease. This study evaluates the synergistic effects of stress-tolerant plant growth-promoting rhizobacteria (PGPR) and reduced chemical fertilizers on tef to enhance growth parameters, yield components, and nutrient content. The experiment was conducted using a completely randomized design with three replications. Stress-tolerant PGPR was applied individually and in consortium with half the recommended dose of chemical fertilizers at the seeding and flowering stages to facilitate effective root colonization and growth. Variance analysis revealed significant improvements in various growth and yield parameters of the Dukem tef variety (Dz-01-974) due to the integrated application of the PGPR consortium and reduced chemical fertilizers increased grain yield per plant by 32% (5.25 g vs 3.98 g control;  $P < 0.01$ ) and shoot dry weight by 28% (10.4 g vs 8.1 g control;  $P < 0.01$ ). Additionally, the grain nutrient content significantly ( $p < 0.05$ ) improved, with phosphate at 3.83%, nitrogen at 1.99%, and calcium at 0.18% over the control. Integrating PGPR with reduced chemical fertilizers offers a promising strategy for improving tef growth and yield, as well as reducing dependency on chemical inputs. This approach has the potential to promote more sustainable agricultural practices, improve the nutritional content of tef, and lower production costs for farmers. Further long-term studies are necessary to validate these findings and explore the broader implications for soil health and crop productivity.

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### Introduction

This study investigates the integrated application of stress-tolerant plant growth-promoting rhizobacteria (PGPR) and reduced doses of chemical fertilizers to enhance the growth and yield of tef [*Eragrostis tef* (Zucc.) Trotter]. By evaluating their synergistic effects, this research aims to develop sustainable strategies for improving tef productivity while addressing challenges associated with environmental stress and excessive reliance on chemical fertilizer.

Tef is a staple crop in Ethiopia, serving as the primary food source for approximately 70

million people and contributing significantly to the country's agricultural economy (Bekana *et al.*, 2022). As a dietary staple, tef is highly valued for its nutritional profile, including high protein content, gluten-free nature, and rich reserves of essential nutrients such as iron (Fe), calcium (Ca), magnesium (Mg), and dietary fiber, making it suitable for people with celiac disease or gluten sensitive (Woldeyohannes *et al.*, 2022; Gebru *et al.*, 2020). Alebachew (2023) and Alias *et al.* (2022) emphasize that tef is a dietary staple for millions of Ethiopians, providing energy and nutrients essential for

human health. Woldeyohannes *et al.* (2022) and Desalegn (2017) have highlighted that tef is rich in essential nutrients such as iron, calcium, magnesium, and dietary fiber. In addition, tef is a versatile crop and can be used to prepare various traditional Ethiopian dishes such as injera, a spongy flat bread that is a staple in the region (Homem *et al.*, 2022).

Tef's adaptability to diverse agroecological zones and resilience to environmental stressors make it a vital crop in Ethiopian agriculture. However, abiotic stresses such as drought, salinity, and nutrient deficiencies pose significant challenges to its production (Assefa *et al.*, 2022). Abraha (2016) and Belete (2020) have documented tef's adaptability to marginal lands, low-input farming systems, and water-stressed environments. Environmental stresses such as drought, salinity, soil nutrient deficiencies, and pest/disease pressures can significantly affect tef's growth, development, and yield. Assefa *et al.* (2022) highlighted the challenges posed by these stressors and their implications for tef cultivation.

Ethiopian farmers have increasingly relied on synthetic inputs, such as chemical fertilizers and pesticides, to boost tef production and productivity. While these inputs have contributed to yield improvements, their overuse has raised concerns about environmental pollution, soil degradation, and reduced soil fertility over time (Pahalvi *et al.*, 2021). Prakash (2023) highlights that long-term dependence on synthetic fertilizers diminishes soil health and disrupts beneficial microbial communities, ultimately threatening sustainable agricultural practices. To address these challenges, integrating sustainable alternatives such as PGPR with reduced doses of chemical fertilizer offers a promising pathway. As demonstrated in the rice-PGPR system, reducing chemical fertilizer use by 50% while maintaining yield aligns with UN Sustainable Development Goals (SDG) 2 and 13 (Kobua *et al.*, 2021). PGPR have emerged as promising biofertilizers due to their ability to promote plant growth and enhance stress tolerance (Etesami and Maheshwari, 2018). They exert their beneficial effects on plants through various mechanisms, including nutrient mobilization, nitrogen fixation, production of growth-promoting substances,

control of pathogens, induction of systemic resistance, and enhancement of stress tolerance. PGPR improves the availability of nutrients for plants, reducing dependence on synthetic fertilizers. Hakim *et al.* (2021) demonstrated the ability of PGPR strains, such as *Bacillus* and *Pseudomonas* species, to improve P uptake and promote root growth, resulting in increased crop yields. Moreover, their ability to produce phytohormones and stimulate root growth contributes to overall plant vigor and productivity (Hol *et al.*, 2014).

This study aims to address the existing knowledge gap by investigating the integrated application of stress-tolerant PGPR and a reduced dose of chemical fertilizers to enhance the growth and yield of tef. It evaluates the synergistic effects of different combinations of PGPR inoculants and a half-dose of chemical fertilizers on tef growth parameters, yield components, and overall productivity. The findings are anticipated to provide valuable insights into sustainable tef cultivation practices, offering practical solutions to improve crop resilience and productivity in stress-prone environments while reducing environmental impacts and mitigating risks to human health.

## Methodology

### Study area

The experimental trial was conducted at the Debrezeit Agricultural Research Centre (DZARC) in the Oromia National Regional State of Ethiopia in 2020. The site is located at 08° 44' N latitude and 38° 58' E longitude, with an altitude ranging from 1860 to 1900 meters above sea level, approximately 47 km southeast of Addis Ababa. Soil for the experiment was collected from a local farm that had been under continuous tef cultivation for several years. The experimental soil had a silt loam texture composed of 14% clay, 32% sand, and 54% silt. Its organic carbon content was measured at 1.26%, which is considered low based on Roy *et al.* (2006). The available phosphorus (P) content was below 3 mg/kg, classifying it as low according to Olsen *et al.* (1954). The soil pH was recorded at 6.96, which falls within the optimal range (4- 8) for tef cultivation. The total nitrogen (N) content was 0.12%, classified as medium

according to Havlin *et al.* (1999). To prepare the soil for the experiment, it was sieved to remove debris and sterilized using an autoclave to eliminate potential microbial contaminants. Subsequently, surface-sterilized plastic pots (12 cm × 12 cm) were filled with 500 g of the prepared sterilized soil.

### Experimental design and treatments

The experiment followed a completely randomized design (CRD) with seven treatments and 42 replications. PGPR strains were selected based on physiological and biochemical evaluations, as summarized in Tables 1 and 2. The criteria for selecting potential PGPR strains included plant growth-promoting properties such as phosphate solubilization (PS), indole-3-acetic acid (IAA) production, nitrogen fixation (NF) ability, and siderophore production. Biocontrol properties were also considered, including the production of hydrogen cyanide (HCN), which inhibits plant pathogens, and the ability to produce antimicrobial compounds that suppress harmful microorganisms. Additionally, tolerance to abiotic stress was evaluated, focusing on the production of exopolysaccharides (EPS) and the

ability of the strains to thrive under varying pH levels, salt conditions, and temperature extremes. The seed vigor index (SVI) was examined to ensure that the PGPR strains promote efficient seed germination, root development, and early plant establishment. Furthermore, metabolic versatility, including the ability to utilize various carbohydrates (e.g., glucose, fructose, sucrose), produce organic acids (e.g., citric acid, acetic acid), and synthesize amino acids (e.g., L-alanine, L-arginine), was assessed to determine the adaptability and nutritional benefits of the strains.

### PGPR strain compatibility test

The compatibility of the selected PGPR strains was assessed to ensure they could coexist without antagonistic interactions. The methodology described by Nikam *et al.* (2007) was followed with slight modifications. Bacterial cultures were streaked on nutrient agar plates, with one bacterial strain at the center and others radiating outward. After 48 hours of incubation at 30°C, inhibition zones were checked. Strains that showed no inhibition zones were selected for use in the consortium.

**Table 1.** Potential PGPR strains selected for greenhouse experimental trial.

PGPR strains	PGPR properties									Seed germination status
	Plant growth-promoting			Biocontrol			Abiotic stress tolerance			
	PS	IAA	NF	Pro	HCN	EPS	SL%	pH	TP	
<i>Serretia marcescens</i> ss <i>marcescen</i>	+++	+++	+	+++	+	++	5	4,5,7&9	40	530
<i>Pseudomonas fluorescent</i> biotype G	+++	+++	++	++	+++	+++	10	5,7, &9	30	470
<i>Enterobacter cloacae</i> ss <i>disolvens</i>	++++	++	+++	+++	+	+++	15	5,7	40	540

Notes: PSI= Phosphate solubilization (mm), IAA= Indole acetic acid (+=colorless (1), +=light pink color formation (2) & +++=pink color formation (3), NF= Nitrogen fixation (+=light yellow color formation (1), +=yellow color formation (2) & +++=orange color formation (3). Pro= Protease enzymes synthesis (+=small area of clearance (1), +=medium area of clearance (2), & +++=large area of clearance (3), HCN= Hydrogen cyanide (+= colorless (1), +=light pink color formation (2) & +++=pink color formation (3) EPS= Exopolysaccharide (+=low precipitation (1), +=moderate precipitation (2) & +++=high precipitate (3); SL= Salinity tolerance level, pH= Potential of hydrogen, TP= Temperature, += Positive for carbon utilization and -= Negative for carbon utilization

**Table 2.** Carbohydrates, organic acid, and amino acid potential of PGPR strains selected for greenhouse experimental trial.

PGPR strains	Carbohydrates								Organic acid		Amino acid		
	$\alpha$ -D-glucose	D-fructose	Galactose	Sucrose	Mannitol	Mannose	Trehalose	Raffinose	Lactic acid	Citric acid	Acetic acid	L-alanine	L-Arginine
<i>S. marcescens</i> ss <i>marcescens</i>	+	+	+	+	+	+	+	-	+	+	-	+	-
<i>P. fluorescent</i> biotype G	+	+	-	+	-	+	+	-	+	+	-	+	+
<i>E. cloacae</i> ss <i>disolvens</i>	-	+	+	+	-	+	+	+	-	+	-	+	-

### Viability of PGPR strains

The viability of the PGPR strains in the presence of chemical fertilizers was tested to determine their survival in fertilizer-amended soil. Solutions of urea and DAP fertilizers at concentrations of 50% and 100% were sterilized and incorporated into the nutrient broth before inoculating with bacterial strains. Bacterial colony-forming units (CFU) were estimated using a turbidimeter, ranging from  $10^6$  to  $10^8$  CFU/mL. Individuals or consortia of PGP bacteria were inoculated in a sterilized broth containing the assigned concentration of chemical fertilizers and incubated at 30°C for 48 hours. The extract of uninoculated nutrient broth was used as a control for comparison.

### Seed surface sterilization and inoculation

Tef seeds were surface sterilized by immersing them in 70% ethanol for 3 minutes, followed by a 5-minute immersion in 1% sodium hypochlorite. The seeds were then rinsed five times with sterile distilled water. Bacterial isolates were grown in Luria Bertani (LB) broth and incubated at 28°C for 24 hours in a rotary shaker. After centrifugation at 5000 rpm for 10 minutes, the bacterial pellets were re-suspended in phosphate-buffered saline (PBS), and the concentration was adjusted to  $OD_{500} = 0.5$  to 1.5, using ELISA spectrometry. For each treatment, surface sterilized seeds were inoculated with either an individual or a consortium of stress-tolerant PGP bacterial species. The inoculated seeds were then shade-dried before sowing.

### Treatment combination

Two tef varieties, Magna (DZ-01-1960) and Dukem (DZ-01-974), were used in the study. The treatments include a control (T1), half a dose of chemical fertilizer (T2), a full dose of chemical fertilizer (T3), and the application of PGPR in combination with half a dose of chemical fertilizer (Table 3). These PGPR treatments consist of *Serratia marcescens* ss *marcescens* (T4), *Enterobacter cloacae* ss *dissolvens* (T5), *Pseudomonas fluorescens* biotype G (T6), and a consortium of *S. marcescens*, *E. cloacae*, and *P. fluorescens* combined with half a dose of chemical fertilizer

(T7). This structure allows for assessing individual and combined effects of PGPR and fertilizers on tef growth and yield.

### Planting and chemical fertilizer applications

Four seeds from each treatment were planted in each of the 42 pots. Phosphate fertilizer in the form of di-ammonium phosphate (DAP) was applied at a rate of 46 kg/ha (0.7 g/pot) at planting, while nitrogen fertilizer in the form of urea was applied at 60 kg/ha (0.9 g/pot), split into two doses: 0.45 g/pot at planting and 0.45 g/pot at the mid-tillering stage.

### Seedling growth and bacterial re-inoculation

To reduce competition among seedlings and ensure optimal growth, the number of seedlings per pot was reduced to five days after emergence. Regular watering with sterile distilled water was maintained to ensure proper soil moisture until physiological maturity.

A second bacterial inoculation was performed seven days after seedling emergence, with 5 mL of bacterial inoculum ( $10^6$ -  $10^8$  CFU/mL) applied per pot. A third inoculation was done 15 days after seedling emergence using the same concentration as the previous application.

### Greenhouse data measurement

At physiological maturity, plant growth, yield, yield-related components, and other data were collected before and after harvest according to the tef descriptors. Plant height (PH) was measured at physiological maturity from the ground level to the tip of the panicle from five randomly selected tef varieties in each plot. Panicle length (PL) is the panicle length from the node where the first panicle branches emerge to the tip of the panicle, which was determined from an average of five randomly selected tef varieties per plot. The number of fertile tillers (NFT) was determined by counting the tillers. Shoot dry weight (SDW) was calculated as above-ground total (shoot plus grain) weight in kilograms. Root dry weight (RDW) was calculated as the total weight of the below-ground in kilograms. Grain yield per plant (GYPP) was measured by harvesting the crop from each pot (g/pot).  $GYPP = (\text{Grain yield per pot (g)} \times 10,000) / \text{pot size (m}^2)$

**Table 3.** Treatment combination.

Factors	Levels	Description
Variety	2	1 Magna
		2 Dukem
Treatment	7	T1 Control
		T2 Half a dose of chemical fertilizer
		T3 Full dose chemical fertilizer
		T4 <i>S. marcescens</i> ss <i>marcescens</i> + ½ dose chemical fertilizer
		T5 <i>E. cloacae</i> ss <i>dissolvens</i> + 1/2 dose chemical fertilizer
		T6 <i>P. fluorescens</i> biotype G + 1/2 dose chemical fertilizer
		T7 <i>S. marcescens</i> ss <i>marcescens</i> + <i>E. cloacae</i> ss <i>dissolvens</i> + <i>P. fluorescens</i> biotype G + ½ dose chemical fertilizer

### Tef grain nutrient analysis

The grain nutrient analysis was carried out according to the methodology described by Miyazawa *et al.* (1999). Tef seed powder (100g) was prepared from each treatment and analyzed for macro- and micronutrient content. The N concentration was determined by means of complete digestion in concentrated H<sub>2</sub>SO<sub>4</sub> and subsequent distillation using the micro-Kjeldahl method. Total K and P were determined using a flame photometer and metavanadate colorimetry,

respectively. Total Ca, Mg, and Zn contents in grain were determined using an inductively coupled plasma atomic emission spectrometer.

### Methods of data analysis

Data were analyzed using R software (version 4.2) to assess the effects of the treatments on plant growth and yield parameters. Analysis of variance (ANOVA) was conducted to assess treatment effects, followed by Tukey's HSD post-hoc tests ( $\alpha = 0.05$ ) for post-hoc comparisons. The least significant difference (LSD) was used to compare individual treatment means, and statistical significance was set at  $p < 0.05$ .

### Result

#### Variance analysis

The two-way analysis of variance (ANOVA) showed that in Table 4. PH, PL, SDW, RDW, and GYPP were significantly ( $P \leq 0.001$ ) improved by treatment; GYPP was also significantly ( $P < 0.001$ ) enhanced by tef variety. Whereas treatment \* variety interaction (Fig. 1) significantly ( $P \leq 0.001$ ) improved PH, PL, and GYPP (Tables 5 and 6).

**Table 4.** Variance analysis of the main and infraction effect of two factors.

SOV	DF	Growth, yield, and yield-related parameters					
		PH	PL	NFT	SDW	RDW	GYPP
TM	6	832.8***	366.8***	0.93 <sup>NS</sup>	4.95***	0.13**	12.7***
VT	1	106.9 <sup>NS</sup>	0.9 <sup>NS</sup>	0.60 <sup>NS</sup>	0.06 <sup>NS</sup>	0.08 <sup>NS</sup>	0.51**
TM: VT	6	109.6*	62.4**	0.38 <sup>NS</sup>	0.38***	0.04 <sup>NS</sup>	1.65***
Error	28	37.3	12.3	0.43	0.02	0.03	0.05

Notes: SOV=source of variation, DF= Degree of freedom, TM= Treatment, VT=variety, PH= Plant height, PL= Panicle length, NFT= Number of fertile tillers, SDW= Shoot dry weight, RDW= Root dry weight, GYPP= Grain yield per plant, \*, \*\*, \*\*\*: statistically significant at  $P \leq 0.05$ ,  $P \leq 0.01$ , and  $P \leq 0.001$  probability level, respectively and NS: not significant.

**Table 5.** Effect of treatment on tef variety growth- and growth-related parameters.

Treatment	Tef growth-promoting traits					
	PH		PL		N FT	
	Magna	Dukem	Magna	Dukem	Magna	Dukem
Control	146.3 <sup>d</sup>	156.3 <sup>d</sup>	35.3 <sup>d</sup>	40.7 <sup>d</sup>	3 <sup>d</sup>	2 <sup>d</sup>
50%	168.1 <sup>c</sup>	169.2 <sup>c</sup>	30.6 <sup>c</sup>	32.4 <sup>c</sup>	2.1 <sup>c</sup>	2.4 <sup>c</sup>
100% NP	179.7 <sup>b</sup>	176.3 <sup>b</sup>	53.0 <sup>b</sup>	53.0 <sup>b</sup>	3.3 <sup>b</sup>	3.3 <sup>b</sup>
<i>S. ss marcescens</i> + 1/2 dose NP	177.7 <sup>b</sup>	177.3 <sup>b</sup>	49.7 <sup>b</sup>	50.7 <sup>b</sup>	3.3 <sup>b</sup>	3.0 <sup>b</sup>
<i>P. fluorescens</i> biotype G + 1/2 dose NP	181.0 <sup>b</sup>	181.7 <sup>b</sup>	56.7 <sup>ab</sup>	49.0 <sup>b</sup>	3.7 <sup>a</sup>	3.3 <sup>b</sup>
<i>E. cloacae</i> ss <i>dissolvens</i> + 1/2 dose NP	173.0 <sup>b</sup>	181.0 <sup>b</sup>	53.7 <sup>b</sup>	55.0 <sup>ab</sup>	3.3 <sup>b</sup>	3.0 <sup>b</sup>
Bacteria consortium + 1/2 dose NP	185.7 <sup>a</sup>	187.3 <sup>a</sup>	60.0 <sup>a</sup>	61.7 <sup>a</sup>	3.3 <sup>b</sup>	4.0 <sup>a</sup>
LSD (0.05) %	7.23	6.88	5.45	7.53	1.05	0.73

Notes: PH= Plant height, PL= Panicle length, NFT= Number of fertile tillers, NP= Nitrogen & phosphate fertilizer, and different letters indicate significant differences at  $P < 0.05$  according to the LSD test.

**Table 6.** Effect of treatment on tef variety, yield, and yield-related parameters.

Treatments	Tef yield and yield-related traits					
	SDW		RDW		GYPP	
	Magna	Dukem	Magna	Dukem	Magna	Dukem
Control	5.8d <sup>c</sup>	6.2d <sup>c</sup>	0.73 <sup>d</sup>	0.93 <sup>d</sup>	1.83 <sup>d</sup>	1.57 <sup>d</sup>
50% NP	6.5 <sup>d</sup>	6.9 <sup>d</sup>	0.69 <sup>de</sup>	0.77 <sup>d</sup>	1.33 <sup>c</sup>	1.40 <sup>c</sup>
100 % NP	8.1 <sup>b</sup>	8.1 <sup>b</sup>	0.94 <sup>c</sup>	0.97 <sup>c</sup>	3.0 <sup>c</sup>	2.01 <sup>c</sup>
<i>S. ss marcescens</i> + ½ dose NP	7.9 <sup>c</sup>	7.3 <sup>c</sup>	0.87 <sup>c</sup>	0.81 <sup>c</sup>	2.93 <sup>c</sup>	2.4 <sup>c</sup>
<i>P. fluorescens biotype G</i> + ½ dose NP	7.2 <sup>c</sup>	7.7 <sup>c</sup>	0.88 <sup>c</sup>	0.91 <sup>c</sup>	3.95 <sup>b</sup>	4.09 <sup>b</sup>
<i>E. cloacae ss dissolvens</i> + ½ dose NP	8.7 <sup>b</sup>	8.2 <sup>b</sup>	1.27 <sup>b</sup>	1.67 <sup>b</sup>	2.92 <sup>c</sup>	4.73 <sup>b</sup>
Bacteria consortium + ½ dose NP	9.99 <sup>a</sup>	10.4 <sup>a</sup>	2.60 <sup>a</sup>	2.91 <sup>a</sup>	4.83 <sup>a</sup>	5.25 <sup>a</sup>
LSD (0.05) %	0.79	0.25	0.76	0.85	0.57	0.96

Notes: NP= Nitrogen and phosphorus fertilizer, SDW= Shoot dry weight, RDW= Root dry weight, GYPP= Grain yield per plant. Different letters indicate significant differences at  $P \leq 0.05$  according to the LSD test.

**Table 7.** Variance of treatment and variety on tef grain nutrient content.

S.O.V	D.F	N %	P %	K %	Mg %	Ca %	Zn %	Fe %
TM	8	0.04 **	1.72 ***	0.002 <sup>NS</sup>	0.002 <sup>NS</sup>	0.004**	0.0001 <sup>NS</sup>	0.0003 <sup>NS</sup>
VT	1	0.03*	0.27 <sup>NS</sup>	0.01*	0.003*	0.0024 <sup>NS</sup>	0.0002*	0.01*
Error	8	0.003	0.24	0.001	0.0001	0.0003	0.0001	0.0004

Notes: S.O.V= Source of variance, D.F= Degree of freedom, TM= Treatment, VT= Variety, N= Nitrogen, P= Phosphate, K= Potassium, Mg= Magnesium, Ca= Calcium, Zn= Zinc; Fe= Iron, \*, \*\*, \*\*\*: statistically significant at  $P \leq 0.05$ ,  $P \leq 0.01$ , and  $P \leq 0.001$  probability level, respectively; NS: Not significant.

### Tef variety grain nutrients content

The experimental data in Table 8 showed that inoculation of either individual or PGP bacterial consortium with a half dose of chemical fertilizer significantly improved tef grain nitrogen and phosphate over the uninoculated treatment. The maximum grains N (1.99%) and P (3.83%) were recorded from a variety inoculated with PGP bacterial consortium and half dose of chemical fertilizer. Similarly, grain Ca uptake was significantly improved by inoculating *Pseudomonas fluorescens* biotype G and PGP bacterial consortium with a half dose of chemical fertilizer over the uninoculated treatment. The maximum grain Ca (0.18%) was recorded from

the variety inoculated by the PGPR consortium with a half dose of chemical fertilizer.

### Correlation of grain yield among other traits

The correlation analysis presented in the scatterplot matrix (Fig. 1) highlights the relationships between grain yield per plant (GYPP) and several agronomic traits. Notably, GYPP exhibits a strong positive correlation with plant height (PH) ( $r = 0.85$ ), panicle length (PL) ( $r = 0.73$ ), and shoot dry weight (SDW) ( $r = 0.81$ ), all of which are statistically significant at the  $p < 0.01$  level. Additionally, GYPP shows a moderate correlation with root dry weight (RDW) ( $r = 0.44$ ,  $p < 0.01$ ).

**Table 8.** Effect of treatment on tef variety, grain nutrients content.

Treatment	N %	P %	K %	Mg %	Ca %	Zn %	Fe %
Control	1.23 <sup>c</sup>	0.62 <sup>c</sup>	0.45 <sup>a</sup>	0.10 <sup>a</sup>	0.06 <sup>b</sup>	0.01 <sup>a</sup>	0.03 <sup>a</sup>
50%	1.42 <sup>c</sup>	0.67 <sup>c</sup>	0.44 <sup>a</sup>	0.09 <sup>a</sup>	0.06 <sup>b</sup>	0.00 <sup>a</sup>	0.04 <sup>a</sup>
100 % NP	1.68 <sup>b</sup>	0.33 <sup>c</sup>	0.38 <sup>a</sup>	0.10 <sup>a</sup>	0.04 <sup>b</sup>	0.03 <sup>a</sup>	0.01 <sup>a</sup>
<i>Serratia marcescens ss marcescens</i> + ½ dose NP	1.82 <sup>ab</sup>	2.44 <sup>b</sup>	0.36 <sup>a</sup>	0.10 <sup>a</sup>	0.11 <sup>b</sup>	0.00 <sup>a</sup>	0.02 <sup>a</sup>
<i>Pseudomonas fluorescens biotype G</i> + ½ dose NP	1.87 <sup>ab</sup>	2.78 <sup>ab</sup>	0.44 <sup>a</sup>	0.11 <sup>a</sup>	0.17 <sup>a</sup>	0.05 <sup>a</sup>	0.05 <sup>a</sup>
<i>Enterobacter cloacae ss dissolvens</i> + ½ dose NP	1.89 <sup>a</sup>	3.63 <sup>b</sup>	0.43 <sup>a</sup>	0.13 <sup>a</sup>	0.07 <sup>b</sup>	0.01 <sup>a</sup>	0.05 <sup>a</sup>
Bacteria consortium +½ dose NP	1.99 <sup>a</sup>	3.83 <sup>a</sup>	0.47 <sup>a</sup>	0.13 <sup>a</sup>	0.18 <sup>a</sup>	0.05 <sup>a</sup>	0.06 <sup>a</sup>
LSD (0.05)	0.19	1.20	0.10	0.05	0.09	0.01	0.10

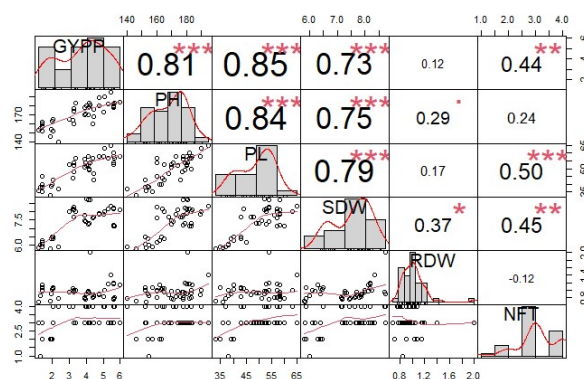
Notes: N= Nitrogen, P= Phosphate, K= Potassium, Mg= Magnesium, Ca= Calcium, Zn= Zinc, Fe= Iron. Different letters indicate significant differences at  $P \leq 0.05$  according to the LSD test.

### Discussion

In this study, the result of the analysis of variance showed that the treatment factor (TM) had significant ( $P \leq 0.001$ ) impacts on plant height, panicle length, shoot dry weight, and

grain yield per plant and grain phosphorus uptake over the uninoculated treatment, and the treatment which received 50% chemical fertilizer. This indicated that the applied treatments were effective in enhancing growth,

yield, and grain phosphorus content. Angulo *et al.* (2020) observed the same trend of PGPR inoculation with chemical fertilizer in improving soil fertility as well as in increasing plant growth, yield, and yield-related parameters. Tef variety (VT) also had a significant effect on GYPP ( $P < 0.01$ ), although it had no significant impact on other parameters such as PH, PL, SDW, and RDW. Moreover, the interaction between treatment and variety significantly improved PH, PL, and GYPP, suggesting that combining specific treatments and tef varieties can lead to superior growth and yield outcomes. The significant interaction indicates that different tef varieties responded differently to the treatments, highlighting the importance of considering both factors for optimizing tef productivity. In terms of growth-related parameters, the analysis indicated that treatments involving the inoculation of plant growth-promoting rhizobacteria (PGPR) combined with a half dose of chemical fertilizer significantly ( $P < 0.05$ ) improved tef growth compared to the control and other treatment groups. Specifically, the maximum growth was observed in *Dukem* (DZ-01-974) inoculated with a consortium of PGPR and half-dose chemical fertilizer, which resulted in the highest values for plant height (PH), panicle length (PL), and number of tillers (NFT). Castanheira *et al.* (2017) tested the same species of bacteria by applying three different strains along with N and P fertilizer supplements to ryegrass (*Lolium sp.*) and observed an improvement in overall plant growth.



**Fig. 1.** Correlation of grain yield per plant with other variables (Own data, 2020)

Adzmi *et al.* (2014) reported the effect of PGP bacteria in combination with chemical fertilizers

on the growth and development of rice. Similarly, Abbas *et al.* (2013) reported the integrated effect of PGP bacteria and chemical fertilizers on the growth of maize. These results suggest that using PGPR, especially with reduced chemical fertilizer application, can enhance growth-related traits in tef, possibly through improved nutrient uptake and better plant health.

Regarding yield and yield-related traits, the application of PGPR, whether singly or in consortium, combined with half-dose chemical fertilizer, resulted in a significant improvement in shoot dry weight (SDW), root dry weight (RDW), and grain yield per plant (GYPP) compared to treatments that only involved chemical fertilizers. The highest SDW, RDW, and GYPP values were achieved when the *Dukem* variety was inoculated with a PGPR consortium and half-dose fertilizer. The results are supported by Saber *et al.* (2012), which also revealed that the co-inoculation of plant growth-promoting rhizobacteria at different levels of chemical fertilizer had a significant role in different yield-related attributes of wheat crops.

The inoculation of *P. fluorescens* biotype G with a half dose of chemical fertilizer also achieved substantial yield improvements, though it was slightly lower than the bacteria consortium. Assainar *et al.* (2018) reported that an adequate combination of microbial inoculants with rock-based fertilizer improved grain yield in maize under glasshouse conditions. The *E. cloacae* ss dissolved with a half dose of chemical fertilizer and showed higher root dry weight than many other treatments, indicating that this specific PGPR strain contributes positively to root growth. Amare *et al.* (2022) demonstrated that combining PGPR and reduced chemical fertilizer significantly increased shoot and root dry weight in tef plants. This synergistic effect suggests that PGPR inoculation enhances nutrient uptake and utilization efficiency, leading to improved biomass accumulation. PGPR may have facilitated the solubilization of essential nutrients in the soil, making them more available to the tef plants. Alias *et al.* (2022) demonstrated that increased absorption of essential nutrients, such as phosphorus (P) and potassium (K), leads to enhanced plant growth, biomass production, and ultimately, higher yields. Additionally, some

PGPRs have the capability to fix atmospheric nitrogen, thereby supplementing the plant's nitrogen requirements. PGPR can also influence various biological processes within the plant, such as hormone production and stress tolerance mechanisms, which could have contributed to enhanced growth and yield. It is an opportunity for poor farmers with low fertilizer investment capacity to optimize the integrated use of PGPR inoculants with a lower rate of chemical fertilizers to achieve higher yields.

The correlation analysis provides important insights into the relationship between plant growth and grain yield per plant (GYPP), offering valuable implications for tef improvement. A strong positive correlation between GYPP and plant height (PH:  $r = 0.85$ ,  $p < 0.01$ ) suggests that taller plants are more likely to achieve higher grain yields. This may be attributed to their enhanced ability to intercept sunlight, which boosts photosynthetic activity, leading to increased biomass production and improved grain filling. Similarly, a significant correlation between GYPP and panicle length (PL:  $r = 0.73$ ,  $p < 0.01$ ) indicates that plants with longer panicles tend to produce more grain, likely due to higher spikelets and improved grain-setting potential. Moreover, shoot dry weight (SDW) exhibits a strong correlation with GYPP ( $r = 0.81$ ,  $p < 0.01$ ), highlighting the importance of vegetative biomass in supporting higher grain yield. Greater shoot biomass may enhance nutrient assimilation and facilitate more efficient carbohydrate partitioning toward grain formation, ultimately contributing to improved yield performance. The treatment also significantly influenced the nutrient content of tef grains. Phosphorus (P) content was notably improved by the treatments at a high level of significance ( $p < 0.001$ ), while nitrogen and calcium contents were significantly influenced at  $p < 0.01$ . Maximum grain phosphorus (3.83%), nitrogen (1.99%), and calcium (0.18%) uptake was observed on a variety inoculated with PGPR consortium with half dose of chemical fertilizer than other treatments. This suggests that the combined application of PGP bacteria and a reduced dose of chemical fertilizers enhances nutrient uptake and accumulation in tef grains. Phosphorus is essential for energy transfer and photosynthesis, and its increased availability can

contribute to better plant health and yield. Nitrogen is crucial for protein synthesis and overall plant growth, indicating that this treatment improves the nutritional value of tef. Ye *et al.* (2019) reported that elements such as N, P, and K are the most essential nutrients for plant growth and development. The VT played an important role in improving the grain nutrient content, with significant effects on N, K, Mg, Zn, and Fe at  $p < 0.05$ . These results suggest that not only the treatment but also the variety of tef can influence the nutrient profile of the grains. For instance, *Dukem* exhibited superior nutrient content in terms of N, K, Mg, Zn, and Fe compared to *Magna*. The findings highlight the importance of combining both varietal selection and treatment strategies to enhance the nutrient content of tef grains, which could contribute to improved nutritional quality for consumers. Higher nutrient uptake in plants could be attributed to the effective translocation of essential nutrients due to better biological nitrogen fixation and mineral solubilization by the introduced PGPR inoculants, which conform to the findings of Mohandas.

## Conclusion

The study demonstrated that the integrated application of stress-tolerant PGPR with a reduced dose of chemical fertilizers significantly enhanced tef variety's growth, yield, and yield-related traits and grain nutrient uptake. The combination of PGPR with half doses of chemical fertilizers resulted in superior plant height, panicle length, shoot dry weight, root dry weight, and grain yield per plant. It also improved the nutritional quality of tef grains, making it a valuable food source compared to other treatments. The superior results achieved with the Bacteria consortium and a half-dose fertilizer highlight its potential as a key strategy for improving tef growth and productivity. Utilizing PGPR alongside reduced chemical fertilizers offers a more sustainable approach to tef crop management. The findings support adopting sustainable agricultural practices that align with environmental and economic goals. Further research could explore additional PGPR combinations, optimize application methods, and assess long-term impacts on soil health and crop productivity.



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## Conflict of interests

The authors declare no conflict of interest.

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