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Research Article

Enhancement of Wheat Yield through Combine Effects of Bio Solution and Bio Enzymes under Low Fertile Soil Condition

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ABSTRACT

Bio-enzymes are the mixture of biological ingredient made up of peels from different fruits, which plays a critical role in catalyzing biochemical processes that transform soil organic matter (SOM) into bioavailable nutrients. Thereby promoting carbon and nutrient cycling and enhancing microbial activity to improve soil quality, fertility and boost the wheat production. Field experiment was conducted at soil research section, Sakrand to examine the efficacy of modified lab made solutions on yield and yield components of two different wheat cultivars (IMDAD 95 and IV2) in Randomized Complete Block design. The results indicates that significant difference was observed in yield and yield components and NPK concentrations. Data suggested that the highest plant height (80.63 cm), thousand grain weight (94.37 g), biomass yield (7.77 kg/m²), grain yield (80.93 kg/acre), spike length (12.44 cm) were detected higher at IMDAD 95 as compare to IV2 among the treatment as compare with control. The results also showed that the highest NPK concentration in leaves were also detected at T7 of IMDAD 95 compare with IV2. The Pearson Correlation data indicates that there was highly significant correlation were observed between the plant height, thousand grain weight, biomass yield, grain yield and spike length and highly non-significant correlation on early maturity days with other treatments. Our results, concluded that bio-enzyme solution significantly increases the physiological parameter of both cultivated varieties as the concentration of BE solution increased. Moreover, IMDAD 95 performed better than IV2 in the bio-enzyme solution. It is suggested that modified bio-enzyme solution should use as a commercial product along with fertilizer to boost the agriculture product and reduces the economy losses in the country.

Keywords: Bio-enzyme, Bio-fertilizer, Organic Waste, Plant Growth, Nutrient Uptake, Wheat Characteristics.



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INTRODUCTION

Wheat (*Triticum aestivum*) is a major staple food crop and one of the most important cereal crops, and considered the highest cultivated and consumed crop globally, with global production reported to reach approximately 700.80 million tons (Al- Myali, 2021). Therefore the significance of wheat lies in its role as the primary dietary staple for populations in over 60 countries. From a nutritional perspective, wheat serves as a vital source of calories for human consumption, owing to its high protein content, which can act as a substitute for animal-based proteins. This makes it particularly beneficial for vegetarian diets, which are associated with lower blood cholesterol levels compared to diets that include meat (Al- Myali, 2021). Considered these facts agriculture serves as the backbone of numerous economies and plays a pivotal role in Pakistan's economy,

contributing 18.9% to the national GDP (Shah *et al.*, 2024). Wheat is the most significant agricultural commodity in Pakistan, cultivated predominantly in arid to semi-arid regions. It accounts for 8.2% of the value added in the agricultural sector. Approximately 80% of farmers grow wheat across 9 million hectares, representing about 40% of the total cultivated area during the winter (Rabi) season (Economic Survey, 2018).

By 2050, the global population is expected to reach approximately 9.97 billion, posing a significant challenge in meeting the rising food demand. Addressing this demand will require an estimated 70% increase in food production (Del Buono, 2021). To achieve higher crop yields and agricultural productivity, the use of chemical fertilizers is a common practice. However, it has been reported that only 30–40% of the applied fertilizers are effectively utilized by plants, with the remainder persisting in the environment. This inefficiency contributes to environmental pollution and poses associated ecological and health hazards (Liu *et al.*, 2023). The excessive use of chemical inputs depletes soil nutrients, disrupts beneficial microbial communities, and accelerates soil degradation, rendering it increasingly infertile. Additionally, hazardous herbicides and insecticides can leach into groundwater during irrigation, contributing to environmental contamination. In light of these challenges, the scientific community is prioritizing the development of sustainable agricultural technologies to improve both the yield and quality of crops. Among these approaches, the application of natural fertilizers, such as bio-enzymes and bio-composites, offers a practical and eco-friendly solution. Soil enriched with bio-enzymes exhibits improved fertility, supports robust plant growth, and helps mitigate insect-borne diseases (Ramprasath *et al.*, 2024).

Bio-enzymes are organic substances that can be produced from various sources, including citrus fruit peels, organic waste, and other biodegradable materials. These complex solutions are typically developed through the fermentation of fresh kitchen waste, such as vegetable and fruit peels (Ramprasath *et al.*, 2024). Bio-enzymes, commonly derived from plant-based fermentation, are characterized as natural, non-toxic, non-flammable, and non-corrosive liquid enzyme formulations (Saini and Vaishnava, 2015). These enzymes serve as critical biochemical components in soil, playing a pivotal role in catalyzing biochemical processes that transform soil organic matter (SOM) into plant-available nutrients. Additionally, they facilitate carbon and nutrient cycling, enhance microbial activity, and improve overall soil fertility (Chuan *et al.*, 2020; Ullah *et al.*, 2021). These metabolic reactions of bio-enzyme in plants are to produce biomass which may suppress due to influence of any metabolic activity during growth stage. In case of wheat plant, it is affected internally through genetic engineering and through external factors like climate and environmental changes (Hassanein *et al.*, 2018; Zaki *et al.*, 2016; Zaki *et al.*, 2012). We hypothesized that the bio-enzyme directly improve the soil physical properties and can be effective to enhanced the crop production keeping this in mind a bio-enzyme solutions were prepared to overcome the wheat production in the local conditions with enhanced the yield and received the higher quantitative and qualitative yield with minimum investment. Novelty, mainly bio-enzyme solution effect was performed and observed on the short duration crop especially vegetables crop, however, we have focused on major crop grown in Sindh province. Therefore, a study was designed to investigate the effect bio-enzyme-solutions prepared under local lab conditions to improve soil fertility and increased the yield and yield components of two wheat cultivars under low fertile soil status.

MATERIALS AND METHODS

Experimental particulars

Soil samples were collected from three depths (0-6, 6-12, 12-30 cm) from the Soil Research Institute Sakrand (26°140 N, 68°268 E) located northern Sindh Province, Pakistan in 2021, (Figure 1). The experiment was conducted at the Soil research Section of Wheat research Institute Sakrand, Sindh, Pakistan. The experiment field was located at humid climate area, characterized by yearly rain is about 128 mm and temperature of 43.3°C. Physiochemical analyses of the soil samples were conducted as follows, Soil particle size was analyzed using a laser diffraction technique (Sochan *et al.*, 2012), The electrical conductivity and pH were measured potentiometrically using a 1:2.5 soil-to-water extract (Sootahar *et al.*, 2019). Soil Organic Matter (SOM) content was determined via wet digestion with sulfuric acid (H₂SO₄) (K₂Cr₂O₇) following the Walkley and Black method (Walkley and Black, 1934), phosphorus content in soil was extracted using 0.5 M NaHCO₃ (pH 8.5) and quantified by spectrophotometer (Sootahar *et al.*, 2022). Available potassium in soil was measured by ammonium acetate (NH₄ O_{Ac}) extraction method and reading was detected on Flame photometer (Knudsen *et al.*, 1982).

The soil used in this study was silty clay, characterized by an alkaline pH, low organic matter (SOM), total nitrogen (N), and available phosphorus (P) content. The soil's physicochemical properties are summarized in (Table 1).

Experimental design

The present study used a complete randomized design that include seven different treatments T1=NP only, T2=NP+0.5% solution "A", T3= NP+ 0.5% Solution "B", T4=NP+0.5% Solution "A" + 0.5% Solution "B", T5= NP+ 1% Solution "A" + 0.5% Solution "B", T6= NP+ 0.5% Solution "A" + 1% Solution "B", T7=NP+ 1% Solution "A"+ 1% Solution "B". respectively, in an area of 3x4 m² plot. There were three replications for each treatment. Wheat seeds were sown on 16 November 2020 with the rate of 60 kg/acre and seeds were bought from wheat research center Sakrand. After harvesting 10 plants were randomly selected and dried in oven at 65 °C for the analysis of nitrogen, phosphorus and potassium concentration in leaves. Harvested plants parts grains, stems were separated and observed the number of tillers/ m², number of spikes/ m², weight of spikes (g/m²) and grain yield(g/m²). Grain yield (ton/acre), straw yield (ton/acre) and biological yield (ton/acre) were examined.

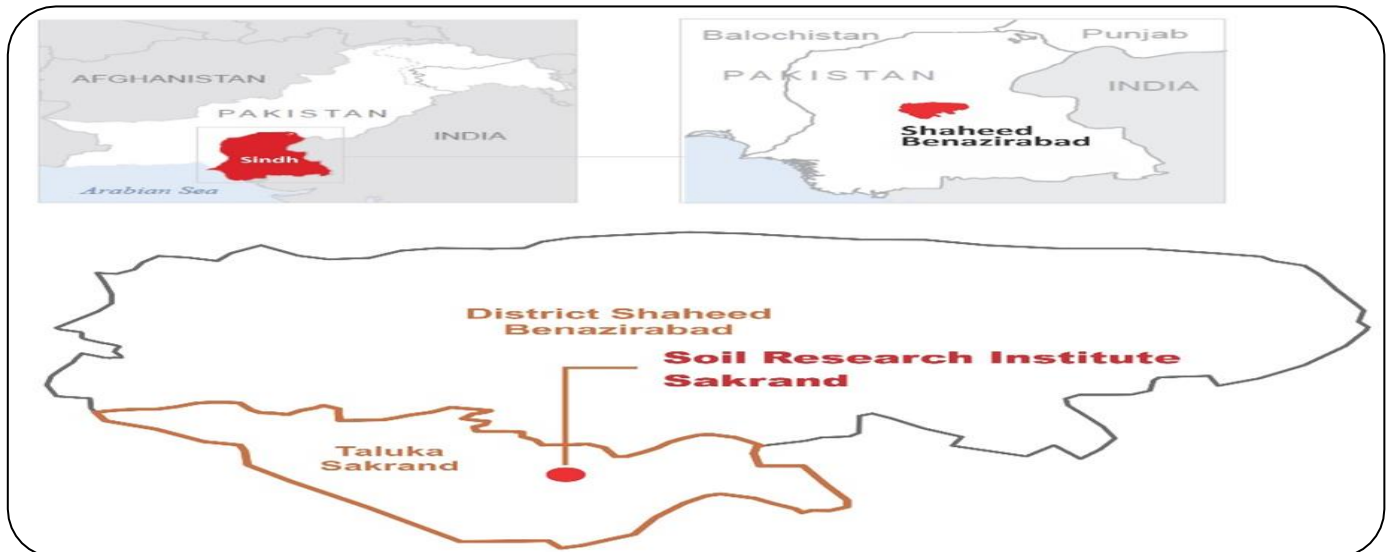


Figure 1: Geographical map of district Sakrand and Soil Research Institute Sakrand.

Table 1: Physiochemical properties of Soil used in this experiment.

Characteristics	0-6cm	6-12 cm	12-18cm
EC (dS m ⁻¹)	1.31	1.27	1.26
pH	8.1	8.0	7.9
T N (%)	0.042	0.043	0.041
K (mg kg ⁻¹)	73	71	69
Available P (mg kg ⁻¹)	4.54	4.44	4.38
Organic matter (%)	0.87	0.84	0.78
Texture	Silty clay Loam		

Note: Electrical Conductivity (EC), Total Nitrogen (TN), Potassium (K), Available Phosphorus (P).

Preparation of Bio-enzyme Solution A and B

Bio-enzyme solution A and B were prepared superlatively at Orchard Nutrient Management Research Institute, Mirpurkhas and applied at Research field of Soil Section Sakrand.

Solution "A": This solution was locally prepared with 1:3:10 of (Jagery, vegetable waste and water) respectively, the mixture was put into air tight bottle and keep for three months. After extracting the solution A, zinc sulphate at the rate of 10 mg/ liter was also added to observe the early maturity of the plant. Whereas, Solution "B" was made from molasses, fruit and vegetable wastes, curd, eggs and farm yard manure. Yeast powder and boil rice were used for fermentation. The mixture was mixed well and left for seven days in dark room with no light passes through. After the extracting the solution B, potassium sulphate 200g, potassium nitrate 300 g, boric acid 20 g, zinc sulphate 30mg and Copper sulphate 10 mg were mixed in the prepared solution B.

Statistical Analysis

The data were subjected to statistical analysis using the analysis of variance (ANOVA) method to evaluate differences among treatment (n=4). Treatment means were compared using the Least Significant Difference (LSD) test at a 5% probability level, performed using the Statistix 24.0 statistical software.

RESULTS

Bio-enzyme and their effect on plant height of two wheat varieties

The effect of Bio-enzyme solution on the plant height of two wheat varieties are presented in (Figure 2). The results revealed that plant height of both varieties significantly increased as compare to control. The data indicate that the plant height of IV2 was significantly increased among the treatments, however observed non-significant at T4, T5, T6 and T7 compare to control, the highest (76.66 cm) plant height was detected at T2 and the lowest (59.66 cm) was observe at T1, similarly, IMDAD 95 were significantly increased between the treatment, however the T6 and T7 were observed non-significant as compare with the other treatments and control. The highest (80.63 cm) plant height of IMDAD 95 were observed at T6 followed by T7 (80.55 cm) and the lowest (60.24 cm) at T1. The data also indicate that IMDAD 95 received the highest plant height as compare with the IV2 among the treatments.

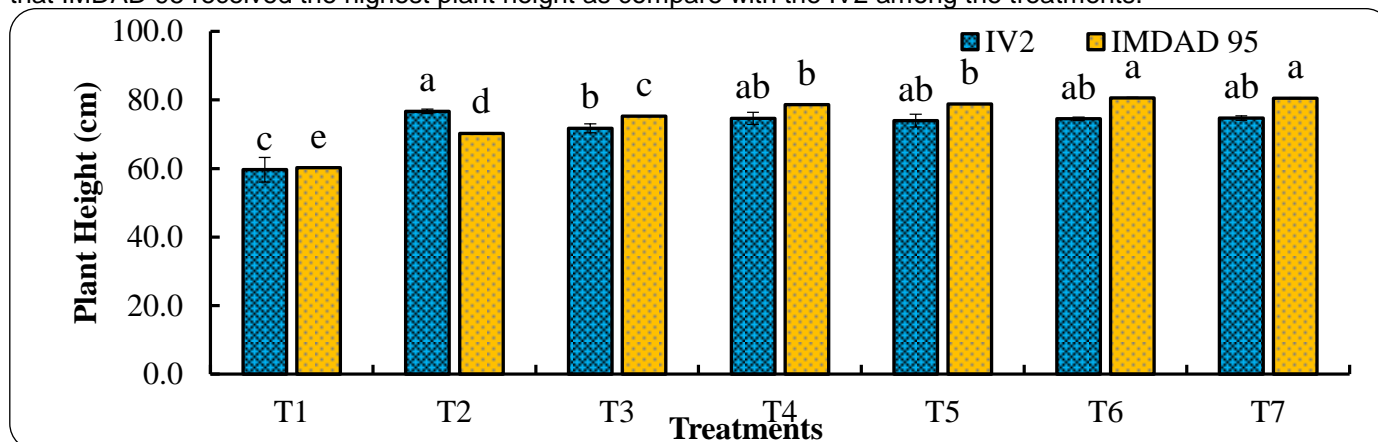


Figure 2: The influence of Bio-enzyme solution T1=NP only, T2=NP+0.5% solution A, T3= NP+ 0.5% Solution B, T4=NP+0.5% Solution A+ 0.5% Solution B, T5= NP+ 1% Solution A+ 0.5% Solution B, T6= NP+ 0.5% Solution A+ 1% Solution B, T7+ NP+ 1% Solution A+ 1% Solution B on Plant height of two wheat Genotypes cultivated during season 2020. The means \pm standard errors are shown (n = 3), bars labeled with the same letter indicate no significant difference at (P = 0.05).

Effect of Bio-enzyme on Thousand Grain Weight of two Wheat Varieties

Thousand Grain weight of two Wheat Varieties after the impact of Bio-enzyme solution were presented in (Figure 3). The data showed that Thousand Grain weight of both varieties IV2 and IMDAD 95 was significantly increased after the implication of bio-enzyme compare with control. The results showed that the IV2 was significantly increased between the treatment however, the highest (88.85 g) thousand grain weight was detected at T6 and the lowest (50.54 g) were observed at T1. Similarly, the thousand grain weight of IMDAD 95 were also significantly increased compare to the control however, observed non-significant between the T4, T5, T6 and T7. The highest (97.37 g) were observed at T6 and followed by (92.88 g) at T7 and the lowest (77.58 g) were detected at T1. Similarly, the thousand grain weight of both varieties were observed non-significant at T6 compare with the other treatments.

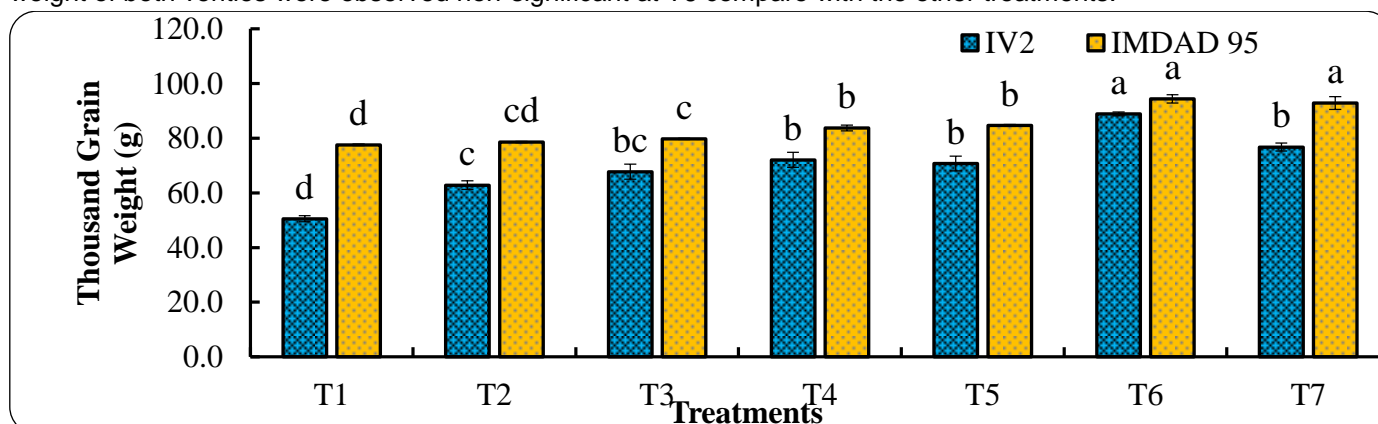


Figure 3: The influence of Bio-enzyme solution T1=NP only, T2=NP+0.5% solution A, T3= NP+ 0.5% Solution B, T4=NP+0.5% Solution A+ 0.5% Solution B, T5= NP+ 1% Solution A+ 0.5% Solution B, T6= NP+ 0.5% Solution A+ 1% Solution B, T7+ NP+ 1% Solution A+ 1% Solution B on thousand Grain Weight of two wheat Genotypes cultivated during season 2020. The means \pm standard errors are shown (n = 3), bars labeled with the same letter indicate no significant difference at (P = 0.05).

Effect of bio-enzyme on bio mass yield of two wheat verities

The effect of bio-enzyme solution on the Bio maa yield of two wheat verities are presented in (Figure 4). The results revealed that biomass yield of both verities was significantly increased as compare to control. The data indicate that the biomass yield of IV2 was significantly increased among the treatments, however observed non-significant at T3, T4 and T6 among the treatment as compare to control, the highest (4.18 kg/m²) biomass yield was detected at T5 and the lowest (1.53 kg/m²) was observe at T1, similarly, IMDAD 95 were significantly increased between the treatment as compare to control. The highest (7.77 kg/m²) biomass yield of IMDAD 95 were observed at T7 followed (7.45 kg/m²) at T6 and the lowest (2.72 kg/m²) at T1. The results showed that IMDAD 95 received the highest biomass yield among the treatments as compare with the IV2 among the treatments.

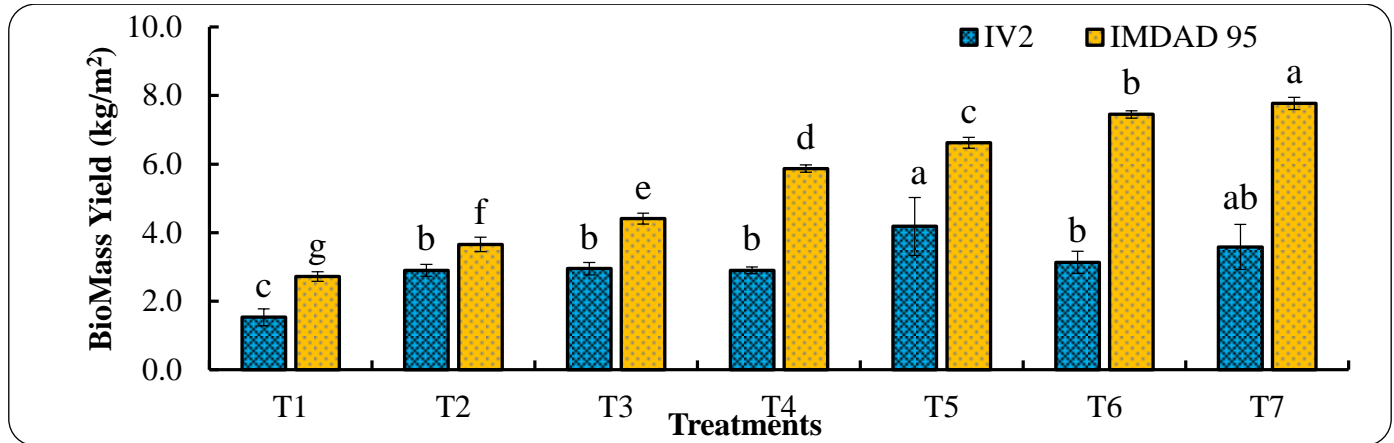


Figure 4: The influence of Bio-enzyme solution T1=NP only, T2=NP+0.5% solution A, T3= NP+ 0.5% Solution B, T4=NP+0.5% Solution A+ 0.5% Solution B, T5= NP+ 1% Solution A+ 0.5% Solution B, T6= NP+ 0.5% Solution A+ 1% Solution B, T7+ NP+ 1% Solution A+ 1% Solution B on Biomass Yield (kg/m²) of two wheat Genotypes cultivated during season 2020. The means ± standard errors are shown (n = 3) bars labeled with the same letter indicate no significant difference at (P = 0.05).

Effect of bio-enzyme on grain yield of two wheat verities

The grain yield of two Wheat verities after the impact of bio-enzyme solution were presented in (Figure 5). The data showed that grain yield of both verities IV2 and IMDAD 95 was significantly increased after the implication of bio-enzyme compare with control. The results showed that the IV2 was significantly increased between the treatment and non-significant at T2 and T3 as compare to control, however, the highest (74.37 kg/acre) grain yield was detected at T5 and the lowest (45.26 kg/acre) were observed at T1. Similarly, the grain yield of IMDAD 95 was also significantly increased compare to the control however, observed non-significant between the T2, T3, T5 and T7 respectively. The highest (83.76 kg/acre) were observed at T6 and the lowest (34.44 kg/acre) were detected at T1. Similarly, the grain yield of both verities was observed non-significant at T1, T2 and T3 compare with the other treatments.

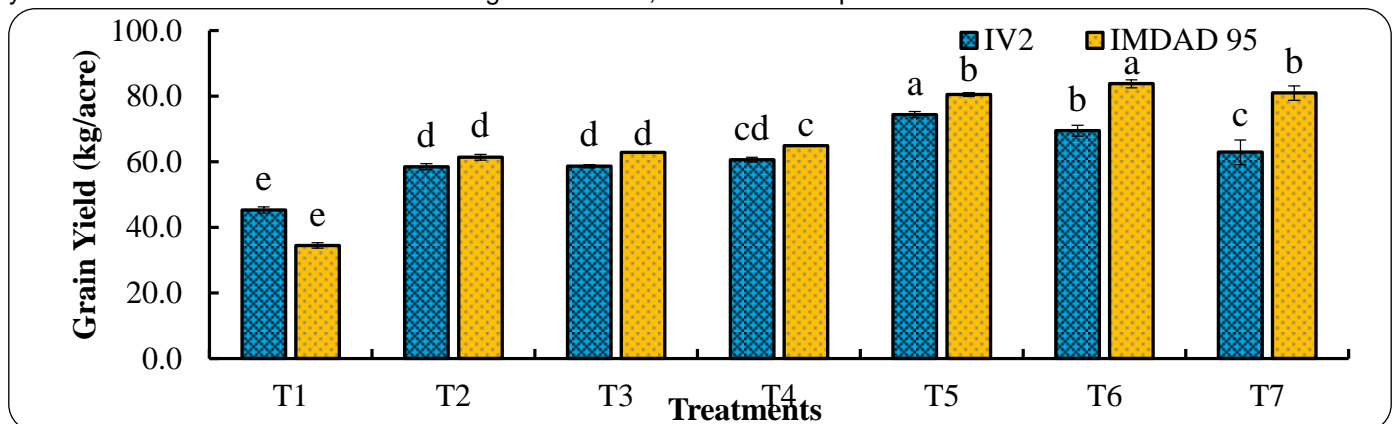


Figure 5: The influence of Bio-enzyme solution T1=NP only, T2=NP+0.5% solution A, T3= NP+ 0.5% Solution B, T4=NP+0.5% Solution A+ 0.5% Solution B, T5= NP+ 1% Solution A+ 0.5% Solution B, T6= NP+ 0.5% Solution A+ 1% Solution B, T7+ NP+ 1% Solution A+ 1% Solution B on Grain Yield (kg/acre) of two wheat Genotypes cultivated during season 2020. The means ± standard errors are shown (n = 3) bars labeled with the same letter indicate no significant difference at P = 0.05.

Effect of bio-enzyme on spike length of two wheat varieties

The effect of bio-enzyme solution on the spike length of two wheat varieties are presented in (Figure 6). The results revealed that spike length of both varieties was significantly increased as compare to T1. The data indicate that the spike length of IV2 was significantly increased among the treatments, however observed non-significant from T2 to T7 as compare to T1, the highest (11.25 cm) spike length was detected at T4 followed by T5 (11.250 cm) respectively and the lowest (9.00 cm) was observe at T1, similarly, IMDAD 95 were significantly increased between the treatment and observed non-significant at T3, T4, T5 and T6 and T7 as compare to T1. The highest (12.44 cm) spike length of IMDAD 95 were observed at T7 followed (12.44 cm) at T6 and the lowest (10.58 cm) at T1. The results showed that IMDAD 95 received the highest spike length among the treatments as compare with the IV2 among the treatments however, observed non-significant from T3 to T7 between the varieties.

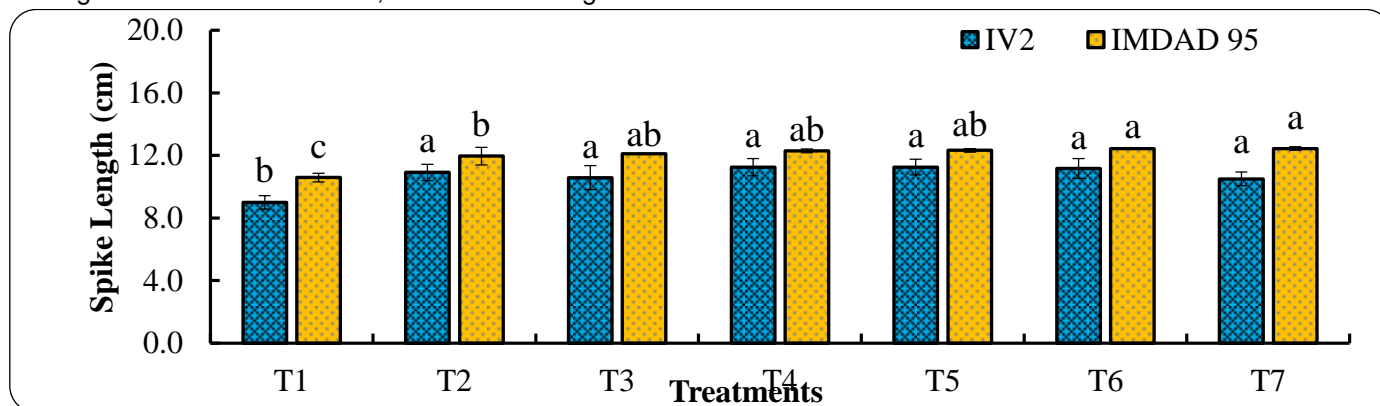


Figure 6: The influence of bio-enzyme solution T1=NP only, T2=NP+0.5% solution A, T3= NP+ 0.5% solution B, T4=NP+0.5% solution A+ 0.5% solution B, T5= NP+ 1% solution A+ 0.5% solution B, T6= NP+ 0.5% solution A+ 1% solution B, T7+ NP+ 1% Solution A+ 1% solution B on spike length (cm) of two wheat genotypes cultivated during season 2020. The means \pm standard errors are shown (n = 3), bars labeled with the same letter indicate no significant difference at P = 0.05.

Effect of bio-enzyme on early maturity days of two wheat varieties

The early maturity days of two Wheat varieties after the impact of bio-enzyme solution were presented in (Figure 7). The data showed that early maturity days of both varieties IV2 and IMDAD 95 was significantly decreased after the implication of bio-enzyme compare with T1. The results showed the IV2 was significantly decreased between the treatment as compare to T1, however, the highest (121.66) early maturity days was detected at T1 and the lowest (106.00) were observed at T7. Similarly, the early maturity days of IMDAD 95 were also significantly decreased and were observed non-significant from T2 to T5 among the treatment as compare to the T1. The highest (124.00) were observed at T1 and the lowest were detected at T7. Similarly, the early maturity days of both varieties was observed non-significant at T1, T2 and T4 compare with the other treatments. Moreover, the early maturity days were observed significantly decreased between the varieties.

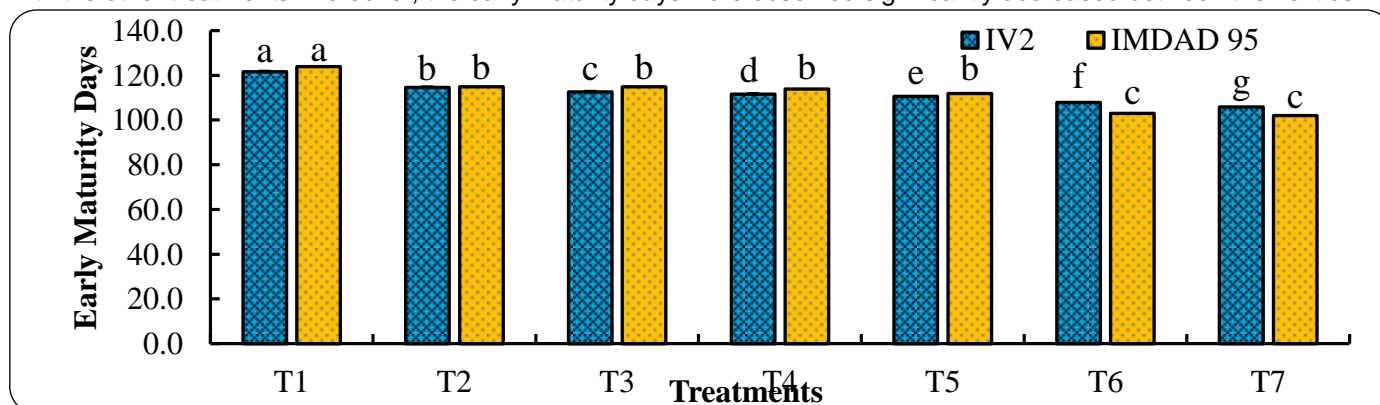


Figure 7: The influence of bio-enzyme solution T1=NP only, T2=NP+0.5% solution A, T3= NP+ 0.5% solution B, T4=NP+0.5% solution A+ 0.5% solution B, T5= NP+ 1% solution A+ 0.5% solution B, T6= NP+ 0.5% solution A+ 1% solution B, T7+ NP+ 1% solution A+ 1% solution B on early maturity days of two wheat genotypes cultivated during season 2020. The means \pm standard errors are shown (n = 3), bars labeled with the same letter indicate no significant difference at P = 0.05.

Impact of bio-enzymes on nutrient uptake of two wheat varieties

Bio-enzyme application along with NP fertilizer effect on the uptake of NPK nutrients are presented in (Table 2). We observed that NPK uptake of both wheat varieties were significantly increased after the application bio-enzyme compare to T1. The results indicated that N, P and K of IV2 was significantly increased the highest N, P and K (2.94%, 0.48%, 2.15 %) were obtained at T7 respectively and the lowest (2.54%, 0.28% and 1.32%) compare with T1 respectively. However, it was also observed that potassium uptake of IV were also non-significant between the T1 and T3. Similarly, we have noticed that N, P and K uptake of IMDAD 95 was also significantly increased as the treatment increased the highest (2.87% N) and (2.24% K) uptake were recorded at T7, and highest P were observed T5 compare to T1. However, it was observed that P was non-significant between T1, T2, T6 and T7 similarly, K was non-significant at T2 and T3 compare with other treatments. The Table also showed that IMDAD 95 has the highest nutrient uptake compare with IV2 on all treatments and T7 received the highest value in IMDAD 95.

Table 2: Influence of bio-enzymes on nutrients uptake of wheat varieties.

Treatments	IV2			IMDAD 95		
	Nitrogen	Phosphorus	Potassium	Nitrogen	Phosphorus	Potassium
	------(%)-----			------(%)-----		
T1	2.54±0.02f	0.28±0.02f	1.32±0.0d	2.52±0.02g	0.25±0.01d	1.31±0.01f
T2	2.60±0.02e	0.31±0.02e	1.37±0.01d	2.58±0.020f	0.27±0.01d	1.35±0.01e
T3	2.64±0.01d	0.35±0.01d	1.42±0.01d	2.63±0.03e	0.31±0.01c	1.38±0.01e
T4	2.67±0.02d	0.37±0.02cd	1.64±0.01c	2.68±0.01d	0.36±0.02b	1.42±0.02d
T5	2.72±0.02c	0.38±0.00c	1.91±0.05b	2.70±0.02c	0.38±0.01a	1.50±0.01c
T6	2.83±0.03b	0.43±0.01b	2.06±0.04a	2.78±0.02b	0.37±0.02ab	1.90±0.02b
T7	2.94±0.02a	0.48±0.01a	2.15±0.04a	2.87±0.01a	0.37±0.02ab	2.24±0.03a

Different treatments of bio-enzymes T1=NP only, T2=NP+0.5% solution A, T3= NP+ 0.5% solution B, T4=NP+0.5% solution A+ 0.5% solution B, T5= NP+ 1% solution A+ 0.5% solution B, T6= NP+ 0.5% solution A+ 1% solution B, T7+ NP+ 1% solution A+ 1% solution B influence on nutrient uptake of wheat.

Pearson correlation between the parameters after bio-enzyme solution implication on wheat varieties

The results of Pearson correlation between the parameter of wheat after the implication of bio-enzyme solution are presented in (Table 3). Data revealed that plant height, thousand grain weight, grain yield and spike length of both wheat varieties observed the highly significant correlation. Similarly, early maturity days were observed highly non-significant plant height, thousand grain weight, grain yield and spike length of both varieties after the application of bio-enzyme solution.

Table 3: Pearson Correlation data between the variables after the implication of Bio-enzyme

Pearson Correlation	P H	G W	BM	GY	SL	EM
P H	1	.626**	.741**	.861**	.681**	-.801**
G W	.626**	1	.768**	.627**	.763**	-.622**
BM	.741**	.768**	1	.793**	.745**	-.657**
GY	.861**	.627**	.793**	1	.658**	-.858**
SL	.681**	.763**	.745**	.658**	1	-.470**
EM	-.801**	-.622**	-.657**	-.858**	-.470**	1

Note: Plant height (P H), grain weight (GW), bio-mass (BM), grain yield (GY), spike length (SL), early maturity (EM).

DISCUSSION

Bio-enzymes and bio stimulants (BSs) are substances or materials distinct from conventional nutrients and pesticides that influence physiological processes in plants to enhance growth and development (Tamburino *et al.*, 2023). These bio-enzymes are recognized as environmentally sustainable and economically viable solutions for optimizing crop productivity. Over the past decade, the market for bio-enzymes and bio stimulants has expanded significantly. However, the identification and development of novel bio-enzyme products with the potential to improve plant growth and enhance nutritional quality have garnered increasing scientific interest (Chaski and Petropoulos, 2022). Considering these points, for the first time, a lab made bio-enzyme solution along with inorganic fertilizers were applied on experimental field to examined the physiological characteristics and nutrient uptake of two distinct wheat varieties. A field trail results of Our study shows that, the physiological characteristics such as: plant height, thousand

grain weight, biomass yield, spike length, and grain yield of two wheat genotypes of IV2 and IMDAD 95 was significantly increased after the application of bio-enzyme along with NP fertilizer, we also examined that IMDAD 95 received the highest plant height compare with IV2 cultivar (Figure 2-7). The better performance of IMDAD 95 compare with other variety could be due to rapid and immediate response of genetic characteristics. Keeping in view the above facts Ramprasath *et al.* (2024) prepare bio-enzyme from mixed fruits and citrus peels they observed plant growth and development was significantly enhanced by the effect of bio-enzyme application. The growth, development, yield and grain quality of chili crops were enhanced when bio-enzyme were applied with various combinations. Furthermore, the application of minimal amounts of fertilizers in combination with bio-enzymes has been shown to enhance plant performance, reduce production costs, and decrease dependency on chemical fertilizers (Manna *et al.*, 2012). Moreover, the effect of bio-enzyme on oxidative stress and biochemical parameters of mung (*Vigna radiate*) was examine and the results showed that the photosynthetic pigments of mung had a positive impact across all the treatment of bio-enzyme, similarly, biochemical content (protein and carbohydrates) in leaves were also significantly increased after the bio-enzyme application (Patidar *et al.*, 2024). In the same way, foliar spray of bio-enzyme on brinjal in a rabi season were demonstrated that the bio-enzyme improved the yield, growth and morphology of the fruits (Jature *et al.*, 2010). The foliar spray of bio-enzyme also significantly increased the yield, quality and of tomato fruit (Ofosu-Anim *et al.*, 2008). Many scientists suggested that bio-enzyme significantly increased the growth and quality of fruit crops. Singh *et al.* (2020) they observed that the physiological characteristics of chick pea significantly increased when bio-enzyme were applied compare with control. Application of bio-enzyme at the 3 ml/L concentration showed the highest growth and yield of rose (Naik *et al.*, 2010). Similarly, foliar spray of seaweed extracted bio-enzyme increased the dry mass, growth and yield of green gram (Pramanik and Bera, 2013). The bacterial biofertilizer also plays a significant role in increasing the levels of sugars and amino acids, which are crucial in various biological processes, particularly cell division and expansion. Enhanced enzyme activity due to biofertilizer leads to the decomposition of organic compounds, releasing nutrients that become readily available for plant uptake. Amino acids, being essential nitrogen sources, contribute to protein and enzyme synthesis and provide energy, promoting vegetative and root growth. Consequently, nutrient translocation from the plant to the leaves increases, resulting in higher NPK concentrations in the leaves (Lateef *et al.*, 2019). The impact of biofertilizer on nitrogen, phosphorus, and potassium is also due to its ability to improve soil physical and chemical properties and fertility, thus enhancing nutrient absorption by plants (Kareem Mustafa and Al-maliki, 2024). Our experiment results showed that NPK concentration in leaves were significantly enhanced by the application of bio-enzyme as the treatment increases along with the inorganic fertilizer. The highest NP and K concentration were observed at T7 treatment compare with the other treatments, meanwhile it was also observed that NPK concentration significantly receiving the highest values at IMDAD 95 compare with IV2 (Table 2). The increasing nutrient concentration to vegetative parts is attributed to converting non-available forms into absorbable ones, aligning with finding by El-Sayed *et al.* (2014) they have noted that bio-fertilizer or bio-enzyme significant role increasing the average nutrient concentration of NP and K in plants. Similar, results were also obtained by Kareem Mustafa and Al-maliki. (2024) they stated that bio-fertilizers enhanced the nutrient concentration in maize leave (N 1.752%, P 0.449% and K. 1.76%).

CONCLUSION

This study concludes that the application of bio-enzymes formulated from mixed fruit and vegetable waste, curd, eggs, and farmyard manure in a 1:3:10 ratio significantly improved plant growth, grain yield, spike length, thousand-grain weight, and biomass yield. Among the varieties tested, IMDAD 95 exhibited superior growth and development compared to IV2. Future research should focus on investigating the physicochemical properties of bio-enzymes and their integration with conventional fertilizer application practices to optimize their efficacy in agricultural systems.

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NOVELTY STATEMENT

This research work provides novelty regarding the application of Bio-enzyme fully prepared with fruit pale and other extra materials to find out the effect on the physiological characteristics of Wheat as well nutrient content.

AUTHOR CONTRIBUTIONS

Mahendar Kumar Sootahar: Writing-original draft, Conceptualization, conducted experiment. **Hafeezullah Babar:** Conceived the Idea, overall management of the work. **Saibrina Sethar:** Analysis and Interpretation of Results, Visualization. **Ambrin Baby Rajput:** Analysis and Interpretation of Results and Review the literature. **Muhammad Aslam Panhwar:** Data Collection and field experiment layout preparation. **Khalid Hussain Khokhar:** Visualization and collection Data. **Abdul Qudoos Abbasi:** Data Analysis and Data collection. **Mukesh Kumar Sootahar:** Reviewed the original draft. All the authors reviewed the results and approved the final version of the manuscript.

COMPETING OF INTEREST

The Authors declare no conflicts of interest to report regarding the present study.

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