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INTERACTIVE POTENTIAL EFFECTS OF MORINGA LEAF EXTRACTS AND PLANT GROWTH PROMOTING RHIZOBACTERIA AGAINST APHID ATTACK ON A NEWLY DEVELOPED WHEAT VARIETY

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Abstract

Wheat is an important agronomic staple crop compared with other cereals. Many factors, including abiotic and biotic stresses, can reduce wheat products, such as attacks by the insect-like aphid. The technologies of moringa leaf extracts (MLE) and plant growth-regulating rhizobacteria for the control of aphid attacks were studied in the current experiment to determine the effectiveness of non-pathogenic bacteria *Pseudomonas fluorescens* (WCS417r) and *Moringa* leaf extracts against aphid infestation in wheat. The investigation was laid out in three replicates arranged under a split-plot design with two factors as a treatment, i.e., MLE and *P. fluorescens* mixed with the soil of wheat seedlings, but MLE was sprayed as foliar application twice, at 20- and 30-days. The aphid-infested and non-infested plants were observed to check the validity of aimed objectives. It was observed that aphid infestation mitigated the plant's novel production due to decreased plant growth, metabolism, and specific physiological pathways. On the other hand, treatments of MLE and bacteria mitigated the devastating effects of aphid infestation and improved plant growth. Plant root and shoot characters were highly improved when compared with untreated plants. Photosynthetic rates were enhanced by 24-41%, and H_2O_2, AsA, phenolics, proline, and MDA were reduced by 25-52% because of applied treatments. All these improvements lead to stable crop production and are highly recommendable for sustainable agricultural practices.

Key words: Wheat, Moringa leaf extract, Rhizobacteria, Aphids, PGPR.

Introduction

Wheat (*Triticum aestivum*) is an essential agronomical staple crop. Compared with other cereals, it has a stable diet compound best for human health, giving protein, vitamins, carbohydrates, and extra calories. Around 700 million tons of wheat is yielded from almost 200 million hectares of land around the globe cultivated with wheat (FAOSTAT, 2021). As one of the critical constraints for the reduction in wheat production, insects and pests may hamper wheat production, leading to about 25% reduced yield every year (Upadhyay & Singh, 2015). Classified wheat varieties were resistant to biotic/abiotic stress (Munns *et al*., 2006). For other stresses, the lost wheat production was estimated at 88.7% in integrated stresses (salinity + water deficit, etc.) (Alharby *et al*., 2020). Biotic stress is notably disastrous ecological stress. Globally, plant development and crop cultivation in cultivated regions have been found to shrink owing to biotic stress (Vimal *et al*., 2017). Several works in literature have been studied revolving around the detrimental impacts of biotic stresses on plant development, crop yields, chlorophyll synthesis, photosynthesis, proteins formation, ionic imbalances, and other metabolic activities in plants occurring due to elevated biotic stress, especially aphid attacks causing the disturbance in life duration (Alharby *et al*., 2020; Wani *et al*., 2020).

It is quite considerable to investigate the ability of plants and bio-pesticides in agricultural integrated pest management (Bansal & Michel, 2015). Green pesticides give out substances naturally and thus leading to efficient pest management and boosting production per unit area (Manzoor *et al*., 2015; Adetunji *et al*., 2020). Biopesticides depicted a significant effect against plant diseases and pests (Jatoi *et al*., 2020; A-Ani *et al*., 2021). Broad-spectrum artificial insecticides are detrimental to human well-being, cause environmental contamination, spoil the food chain, and eradicate valuable insects. Botanical insecticides such as (*Azadirachta indica* A. Juss.) have been proven helpful as bio-pesticide, precisely described (over 400 species) contrarily to insects (Martinez, 2002), yet it is dire demand to explore such innovative bio-pesticides and signify their malignancy level in opposition to insects, birds, fishes and mammals as they are expected to be eco-friendly, economically viable, and possess zero harmful threats to humans and else non-pursued organisms (Tang *et al*., 2002). Various species native to Meliaceae groups like *Tillia americana*, *A. indica*, *Melia volkensii*, and *Azadirachta exelsa* are influential to noctuid caterpillars, *Trichoplusia ni* and *Pseudaletia unipuncta* (Akhtar *et al*., 2008). Before comprehensive illustration, commercialization of these bio-pesticides is not conceivable.

Moringa oleifera (Moringaceae) is a multipurpose plant, helpful as feed for animals, and considered a healthy alternative for curing human diseases (Mahmood *et al*., 2010). The laboratory investigations of moringa usage have been proven quite beneficial. Also, its diverse applications on crop development and physiological growth are an interesting study for researchers and helpful in creating tolerance opposing pests and diseases (Foidl *et al*., 2001). Moringa leaf extracts (MLE) have the potential to reciprocate plant metabolic activities owing to the presence of zeatin, ascorbic acid, phenolic compounds, and vitamin E in it (Isman, 1997). The moringa leaf extracts are excellent water-purifying agents and can be used as natural coagulants, medicinal herbs, biogas fuel, edible vegetables, livestock food, green manuring, and bio-pesticide (Taiz & Zeiger, 2006). Mechanism of cell elongation as well as division is also linked to MLEs (Manzoor *et al*., 2015), employed as a seed-marinating agent for maize and sunflower (Hala *et al*., 2017) and an excellent plant growth-regulating agent (Bakala *et al*., 2021). However, many microbes possibly hold a decisive role in boosting crop production and controlling pests (Aguilar *et al*., 2020a; Aguilar *et al*., 2020b).

Plant growth-regulating rhizobacteria (PGPR) colonize an area of the rhizosphere (Sharma *et al*., 2020; Singh *et al*., 2021) and have efficacy in the enhancement activity of plants to resist bad conditions both biotic (Al-Ani, 2017) and abiotic stress (Lalarukh *et al*., 2022a; Lalarukh *et al*., 2022b). The study aimed to cover the findings of interactive potency of moringa leaf extracts (MLE) and plant growth-promoting rhizobacteria against Aphid attack on a newly developed wheat variety by biochemical tests to measure the efficacy of the identified rhizobacteria in attenuating biotic stress tolerance in wheat in context of sustainable agriculture.

Materials and Methods

Experimental location and design: This investigation was planted in the semi-arid experimental location of Government College Women University Faisalabad (30° to 31.5° N; 73° to 74° E; 184.4 m above sea level). Randomized complete block design (RCBD) was used and replicated thrice.

Soil properties: A textured clay loamy soil (7-10 inches deep) was obtained from a Faisalabad village agricultural site. This soil was air-desiccated before being sieved with a 2 mm sieve. Some soil properties were pH=8.3; organic matter = 0.80%; EC = 5.79 dS m^{-1}; available K = 132 µg/g, total available N = 0.0031 % and available P = 5.93 µg/g.

Seed collection and sterilization: Anaj 17, a well-suited wheat cultivar, was chosen for this study that responds well to environmental stress conditions, as demonstrated in several earlier researches. Seeds were decontaminated with 95% ethanol, then cleansed in a sodium hypochlorite mixture and washed thrice with distilled water.

Rhizobacterial cultivation: Non-pathogenic bacteria *Pseudomonas fluorescens* (WCS417r) were cultivated on King's mixture B (King *et al*., 1954) agar plates having 25 mg L^{-1} rifampicin for 48 hours at 28°C. Before being combined via autoclaved soil, bacteria were recovered and re-dipped in 10 mm $MgSO_4$ at a density of 10^9 colony-forming units (cfu) per ml having optical density (OD660) = 1.0.

Plant and insect material: Wheat seeds were sown and allowed to grow in a clay loam Soil that was autoclaved twice for 20 minutes at 121°C with a 24-hour break. Three-week-old saplings were shifted into pots having identical soil types and left to flourish for another five weeks. A culture of bacteria (10^9 cfu ml^{-1}) was intermixed thoroughly into the soil to an absolute density of 5×10^7 cfu g^{-1} before the transfer of seedlings to the soil, as stated earlier (Pieterse *et al*., 1996). An equivalent volume of 10 mm $MgSO_4$ was applied to the control soil. Plants were raised in a growth chamber with a 16-hour day-light intensity of 300 mol m^{-2} s^{-1} at 20°C temperature/8 hours dark at 15°C temperature cycle with a constant humidity of 70%. The plants were at about 75% FC (75% field capacity). All the plants stayed in the vegetative stage throughout the trial. The species of aphid *Myzus persicae* (Sulzer) was evaluated in the current study. The aphids were tended in the greenhouse at 22–24°C, 50–70% relative humidity, and a 16-h light: 8-h dark photoperiod on Brussels sprout plants (*Brassica oleracea* L.).

Foliar application of Moringa leaves extract: Fresh leaves from a disease-free, mature moringa tree were picked, adequately cleaned, and frozen overnight. Following the procedure of Yasmeen *et al*., (2013), 100 g of leaf sample was blended in 1 L of distilled water (1:10 w/v) for 15 minutes. Moringa leaf extract (MLE) was filtered and diluted 30 times (1:30 v/v) in a laboratory trial before being sprayed over a pot-grown wheat crop with a hand sprayer. Before application, MLE was freshly prepared. In foliar sprays, 0.1% v/v tween-20 was utilized as a surfactant. A few chemical properties of MLE were total chlorophyll=3.79 mg g^{-1} FW, total carotenoids=1.58 mg g^{-1} FW, total phenols=1.68 mg g^{-1} FW, total sugars=338.23 mg g^{-1} FW, nitrogen=14.13 mg g^{-1} DW, phosphorus=2.98 mg g^{-1} DW, potassium=11.97 mg g^{-1} DW, magnesium=3.01 mg g^{-1} DW, calcium=16.8 mg g^{-1} DW, iron=0.398 mg g^{-1} DW, and zinc=0.05 mg g^{-1} DW. MLE was foliar sprayed on wheat plants twice, at 20- and 30-days following planting. During each application, control plants were mock-sprayed twice with pure water. The diluted MLE solutions were sprayed twice at a 10-day interval with the help of a manual sprayer at the rate of 50 mL per plant.

Aphid infestation: One group of plants was provided with 30 aphid nymphs (4 days old) dispersed on six fully developed leaves. The number of aphids was identical to those mentioned in previous findings (De Vos *et al*., 2005; Kusruerczyk *et al*., 2008). Two days after aphid infection, leaves were harvested by choosing three fully grown leaves per treatment containing four to six aphids. Before collection, a fine paintbrush was used to remove aphids and other remains. Non-infested plants were reared the same way as aphid-infested ones and harvested at similar times.

Seed sowing and treatment plant: Ten wheat seeds were sown (2 cm deep) in each pot (23 cm diameter × 27 cm height) with 10 kg of soil. Following the final emergence, saplings were shrunk to 6 plants per pot. Two groups of aphids-infested and non-infested plants were set in four treatments: (i) control [untreated]; (ii) MLE [Alone]; (iii) bacteria [Alone]; (iv) MLE + Bacteria [combined application of MLE and bacteria]

Harvesting and measurement of growth attributes: Plants were harvested eight weeks after germination, and fresh weights of root (RFW) and shoot (SFW), their lengths, and dry weights (RDW, SDW) were assessed immediately. Each treatment included three replicates; each replicating nine leaves picked from three different plants. The sample was instantly immersed in liquified nitrogen for later fresh analysis and kept at -30°C in a biomedical freezer. Three samples from each amendment were oven desiccated for three days at 65°C to record ionic analysis and dry weight values.

Net photosynthesis: Net photosynthesis was noted for three days on three frequently chosen plants out of each amendment on 3rd or 4th fully matured leaves throughout the treatment period. This was done with the IRGA apparatus (CIRAS-2, PP-systems, MA, USA). The observations were taken seven days after MLE was initially sprayed.

Leaf Biochemical Analysis: 50 mg of dry leaves were dissolved in 10 mL of 80% ethanol and filtered out. To quantify osmolytes and non-enzymatic antioxidants like total soluble sugars (TSS) (Dubois *et al*., 1956), total soluble proteins (TSP) (Bradford, 1954), phenolics (Bray & Thorpe, 1954), proline (Bates *et al*., 1973) and ascorbic acid (Azuma *et al*., 1999), the solution was escorted again in 10 mL ethanol with an absolute volume of 20 mL and assessed by spectrophotometer (Model SM1200; Randolph, NJ, USA).

Malondialdehyde and hydrogen peroxide contents: Heath & Packer (1968) method was used to assess lipid peroxidation in chloroplasts, which included estimating malondialdehyde (MDA) concentrations after the thiobarbituric acid reaction. The procedure was followed by Mukherjee & Choudhari (1983) to calculate the hydrogen peroxide (H_2O_2) content. In 10 mL cold acetone, 0.1 g of leaf sample was obtained and centrifuged at 10,000 rpm. The solution was then amalgamated with a 5 mL ammonium mixture and 4 mL titanium reagent. After centrifugation at 10,000 rpm for 5 minutes, the resulting precipitate was transferred to 10 mL of 2N H_2SO_4. The leftover was centrifuged once more to eliminate any suspended fragments. The optical density at 415 nm was measured with the help of a spectrophotometer (UV-2550; Shimadzu, Kyoto, Japan).

Statistical Analysis

In data analysis, the Statistix.8.1 programs performed two-way ANOVA in a completely randomized block design with a split-plot arrangement. At a 5% significant level (p<0.05), LSD test was performed for all pair-wise comparisons. The process of transforming logarithmic data into a near-normal distribution before analyzing was done where necessary. To determine the relationship between variables, percent change was calculated by general percentage calculation given by Bansilal (2017) according to the given formula:

$$\text{Change (\%)} = \left(\frac{\text{Difference between Pi an Po}}{\text{Po}}\right) \times 100$$

Results

The effects of treatments were significant for Shoot/root fresh weight (g) and Shoot/root dry weight (g) of wheat. Treatment MLE+Bacteria was highly significant for the improvement in SFW over control. Application of bacteria and MLE alone also performed significantly better for SFW than control, but MLE+Bacteria was even better than them. In the case of aphid-infested wheat plants, MLE+Bacteria and bacteria alone were statistically similar but differentiated significantly compared to the control. Treatment MLE remained significantly positive over control for RFW in infested plants. It was observed that aphid infestation led to lowered plant vegetative growth, mitigated by given treatments. On average, bacteria alone caused a maximum increase (54%) in RFW over control. However, wheat showed a 34% significant increase in RFW over untreated plants (Table 1). For SDW, MLE+Bacteria remained significantly better than control in biotically stressed wheat. Application of MLE and Bacteria alone also significantly increased RDW and bacteria differed significantly, but MLE did not remain significant for RDW over control. On average, MLE+ bacteria caused a maximum increase (42-55%) in growth parameters compared to the control (Table 1).

For SDW, MLE+Bacteria remained significantly better than control in biotically stressed wheat. Treatments given to both infested and non-infested plants proved significant for Net Photosynthesis (μmol $m^{-2}s^{-1}$), TSP, TSS, significantly decreasing the AsA. Treatment MLE+Bacteria was significantly best for improving the photosynthesis rate and total solutes in the plant over control, followed by bacteria and MLE alone. Also, the same trend was observed in decreasing the antioxidants being physiological attributes of plants on average; bacteria alone caused a maximum increase (34%) in net photosynthetic rate, TSP and TSS over control. For AsA, MLE+Bacteria remained significantly better than the control in stressed wheat (Table 2).

Applying MLE and Bacteria alone also significantly lowered the H_2O_2, MDA, proline and plant phenolics in response to biotic stress and mitigated their adverse effect on wheat growth. The lowest antioxidants were observed in plants treated with bacteria and MLE under aphid infestation. On average, MLE+bacteria caused a maximum decrease (25%) in these parameters compared to the control (Table 3).

Table 1. Influence of MLE and bacterial inoculation on Shoot fresh weight (g), Root fresh weight (g), Shoot dry weight (g), Root dry weight (g), Root and Shoot length (cm) in aphid-infested and non-infested.

	Shoot fresh weight (g)		Root fresh weight (g)		Shoot dry weight (g)		Root dry weight (g)		Root length (cm)		Shoot length (cm)	
	non-infested	aphid-infested	non-infested	aphid-infested	non-infested	aphid-infested	non-infested	aphid-infested	non-infested	aphid-infested	non-infested	aphid-infested
Control	**0.93d**	0.33d	1.10d	0.63c	0.097d	0.057d	0.087d	0.063d	1.40d	0.63d	1.40d	0.97d
MLE	2.27c	1.77b	2.27c	1.63b	0.190c	0.123c	0.253c	0.203b	2.07c	1.67b	1.70c	1.13c
Bacteria	2.90b	2.47a	3.10b	2.57a	0.307b	0.206a	0.343b	0.287a	2.70b	2.40a	2.37b	2.10a
MLE+Bacteria	3.77a	1.52c	4.97a	1.61b	0.467a	0.148b	0.483a	0.184c	3.57a	1.57c	3.17a	1.40b
Mean	2.47	1.52	2.86	1.61	2.65	1.48	2.92	1.84	2.43	1.57	2.16	1.40

Wheat plants; where MLE= Moringa leaf extract, C.V= Coefficient of variance, S.E= Standard error, LSD= least significant difference at p=0.5, **= Highly significant, *= Significant

Table 2. Effect of MLE and Bacterial inoculation on Net photosynthetic rate, TSP, TSS and AsA in aphid-infested and non-infested wheat plants.

Treatment	Main effect of treatments							
	Net Photosynthesis ($\mu mol\ m^{-2}s^{-1}$)		TSP (mg/gFW)		TSS (µmol /g fwt)		AsA (µg/g fwt)	
	Non-infested	Aphid-infested	Non-infested	Aphid-infested	Non-infested	Aphid-infested	Non-infested	Aphid-infested
Control	2.67d	3.33d	1.68d	2.41d	1.30d	2.56c	2.73a	3.34a
MLE	5.33c	4.33c	2.49c	2.80c	2.49c	3.02bc	2.24b	2.85bc
Bacteria	5.43b	5.00b	2.84b	3.08b	2.63b	3.43b	2.00c	2.61c
MLE + Bacteria	6.33a	6.67a	3.33a	4.58a	3.04a	4.53a	1.57d	2.28d
Mean	5.17	5.33	2.58	2.97	2.36	2.86	1.88	3.00

Table 3. Influence of MLE and Bacterial inoculation on H_2O_2, MDA, phenolics, and Proline contents in aphid-infested and non-infested wheat plants.

Treatment	Main effect of treatments							
	H_2O_2 (µmol/g fwt)		MDA ($\mu mol\ g^{-1}$ FW)		Phenolics (mg/g dwt)		Proline (µg/g f.wt)	
	Non-infested	Aphid-infested	Non-infested	Aphid-infested	Non-infested	aphid-infested	Non-infested	aphid-infested
Control	3.95a	3.74a	2.22a	2.63a	2.98a	3.28a	4.11a	5.23a
MLE	3.44ab	3.51bc	2.09ab	2.52ab	2.89ab	2.99b	3.88b	4.90b
Bacteria	3.21b	3.21c	1.79b	2.30b	2.69b	2.79c	3.65bc	4.87b
MLE+Bacteria	2.78c	3.09d	1.46c	1.87c	2.46c	2.56d	2.73c	3.85c
Mean	3.10	3.40	1.87	2.28	2.76	3.06	3.52	4.64

Where MLE= Moringa leaf extract, C.V= Coefficient of variance, S.E= Standard error, LSD = Least significant difference at p=0.5, **= Highly significant, *= Significant

Discussion

Aphids are the major biotic stress-causing agents that challenge agricultural productivity, especially in semi-arid regions. They retard plant growth and development, which results in destructive consequences on plant development and reproduction. Furthermore, it accompanies metabolism initiating reactive oxygen species (ROS) like superoxide (O_2) and hydrogen peroxide (H_2O_2) (Tsai, 2011). Cheeseman (2007) revealed that ROS provokes oxidative stains in plants and damage chlorophyll, DNA structure, and membrane actions. In the current assessment, an aphid attack caused a considerable growth decline, and wheat's final yield was highlighted. The deviation in plant development and outturn was attributed to an elevated quantity of ions and oxidative stress in the form of H_2O_2 and O_2 levels, which influenced photosynthetic efficacy adversely, gaseous exchange, ionic imbalance, and impaired membrane stability, and accordingly, growth inhibition. Biotic stress also causes the toxic consequences of osmotic pressure, the collapse of metabolic operations, narrow energy necessities, and interruption in cell divisions (Desoky *et al*., 2020; El-Yazal *et al*., 2021). Comparable findings were reported whensoever conceivable; plants are flourishing and accepting an antioxidant scheme to diminish devastating consequences led by ROS (Halliwell & Gutteridge, 2020).

In the current study, aphid-infested and non-infested wheat plants were inoculated with rhizobacteria selected as PGPRs along with MLE, where treated plants were differentiated from control plants. The enhanced improvement of wheat plants owing to PGPRs might be due to the emission of various phytohormones in plant root surroundings and strong synchronization in endogenic levels (Mohammed *et al*., 2014; Desoky *et al*., 2019). Plant hormones uphold cellular division and elongation phenomena (Nayak *et al*., 2014; Soumare *et al*., 2021). Also, PGPRs promote root cell elongation and proliferation by synthesizing IAA (Ahmad & Hasnain, 2014). Therefore, it can be inferred from the current investigation that applied bacterial strains termed PGPRs boosted growth, as well as the development of wheat roots, precisely root hairs, which led to enhanced water and nutrients absorption by roots and plant development, kept normal as justified by results of Cassán *et al.*, (2014) study, in which root structure and physiology were upgraded by induction of various microbial metabolites like auxin, gibberellins, and cytokinin, which ameliorate water along with nutrition absorption effectively. This innovation in wheat development could also be accredited to diverse features of trialed strains like IAA and NH_4 synthesis and the solvability of ionic components. Hence, a correlation might be identified among plant growth-regulating domains of water stress-resistant PGPRs with advanced characteristics and wheat biomass, presenting that those characteristics cannot diminish water stress but can regulate growth parameters.

Plant-microbe synergy is a beneficial corporation between microbes and plants (Cheng *et al*., 2007), turning into an effective approach with cost-effectiveness and sustainability for retrieving vegetation and crop productivity. Destruction of plasma membranes and chlorophyll pigments quantity is occurred owing to oxidative stress by surplus synthesis of H_2O_2 and O_2 (Raddy *et al*., 2019). Oxidative stress biomarkers are linked with boosted Na^+ absorption and declined Mg integration, influencing chlorophyll production (Gomes *et al*., 2017). Pattanayak & Tripathy (2011) disclosed that the accretion of minerals lowers pigments formation, leading to retarded activity of certain enzymes like 5-aminolevulinic acid dehydratase, porphyrinogen IX oxidase, and protochlorophyllide oxidoreductase. Chlorophyll contents of stressed plants proved to be a key agent behind plant well-being and photosynthetic capability in stress. Our experimental data depict that all levels of MLE have considerably deflated aphid infestation in different plant growth phases. Likewise, growth attributes also justified a considerable increment in wheat development.

Moringa leaf extracts (MLE) are full of macro (Ca, P, Mg, Na, and K) together with micro (Fe, Mn, Zn, and Cu) constituents that are fundamental plant growth boosters when employed as growth stimulators (Farooq *et al*., 2010). Before sowing amendments, seed marination and plant growth regulators (PGRs) polish germination percentage, boost total productivity and narrow the possibilities of pest invasion in certain plants (Taylor & Harman, 1990). In stress conditions like salinity, PGRs and growth stimulators upsurge the final outturn of several plants. Fuglie (2000) indicated that using leaf extracts of Moringa (MLE) increases the development pace of plants and declines the pest populace. This investigation demonstrates that foliar amendment of MLE has triggered overall development and outcomes by improving dry matter contents (TDM), leaf area duration (LAD), leaf area index (LAI), crop growth rate (CGR), and other attributes. Also, aphid invasion lessens significantly at booting, milking, and heading stages following the exogenous provision of MLE. Diluted MLE upsurged metabolism, boosted emergence and resulted in dynamic plant development (Seed marinated in MLE) in maize, wheat, range grasses, and certain other crops (Iftikhar, 2009).

Enebe & Babalola (2018) studied the considerable positive impact of PGPR on the preservation of photosynthetic components. They ascribe these outcomes to the factor that amendments with PGPRs might have altered cell wall arrangement and physio-biochemical alterations that might have been the possible reason behind the production of proteins and enzymes linked with pigment firmness.

Higher synthesis of auxins and indirect root architecture improvement by PGPRs aims to add increment in water uptake, and nutrient absorption, and movement to vegetative portions for taking part in boosting the quantity of pigment synthesized and also enabling relocation of photosynthetic assimilates in plants (Ahmad & Hasnain, 2014; Mansoora *et al*., 2021). Other than that, the supporting constituents of carotenoids using PGPRs illuminated the double role of bacteria in implying carotenoid shield and restricting oxidation of carotenoids by ROS (Mathivanan *et al*., 2017; Rehman *et al*., 2022). Besides, PS II actions can also interpret a plant's capability to photosynthesize.

In this assessment, the provision of PGPRs (Rhizobacteria and MLE) boosted photosynthetic effectiveness, TSP, and TSS, and the effect of MLE was very effective. Hence, it is vital to elevate the final yield of cereals employing diverse strategies, like seed marination of MLE in maize, wheat, and other grasses, that has fluctuated the growth positively. This method can be comprehended to native crops for extortionate productivity and pest control (Yaseen *et al*., 2021). Pill & Savage (1988) disclosed that seed priming using inorganic salts, sugar beet extracts, and other plant growth promoters upgrades metabolic processes and germination percentage along with the physiological acclimatization of plants. Insecticides are intensely toxic and can lead to growth inhibition and nourishing deterrents in lepidopteron pests (Amjad *et al*., 2021). Habib *et al.* (2015) studied moringa leaf extract (MLE) in integration with neem leaf extracts (NLE) and eucalyptus leaf extracts (ELE). They identified a considerable down curve in aphid invasion in wheat in contrast to their sole amendments, proposing that moringa leaf and root extracts are well suited together with other pest manipulation techniques. Further investigation is needed to affirm the synergistic impacts of MLE with other pesticides, insecticides, and biological control agents.

Conclusions

We can conclude from this study that the application of rhizobacteria and moringa leaf extract is recommended to increase wheat's morphological attributes, which greatly contributes to sustainable agriculture. It was found that treating both factors improved wheat's root and shoot attributes. Moreover, the application of combined treatments of rhizobacteria and moringa leaf extract against aphid attacks causing biotic stress recorded the least mean values of antioxidants leading to cell injury and reduction in plant biomass, consequently decreasing the yield.

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