Biological Forum
an International Journal 17(7): 102-108(2025)
ISSN No. (Print): 0975-1130
ISSN No. (Online): 2249-3239

Antipathogenic Activity of Antagonistic Bacterial Isolates Against *Macrophomina phaseolina* and their Role in Inducing Biochemical Defence in Black Gram under Pot Conditions

Prajapati M.D.[1]*, Patel H.H.[2], Kandoriya P.J.[3], Joshi R.L.[1] and Chaudhari A.K.[4]
[1]Ph.D. Research Scholar, Department of Plant Pathology, NMCA, Navsari, (Gujarat), India.
[2]Ph.D. Research Scholar, Department of Life Science, Aberystwyth University, Wales, UK.
[3]Ph.D. Research Scholar, Department of Agricultural Microbiology, NMCA, Navsari, (Gujarat), India.
[4]Junior Research Fellow, Anand Agricultural University, Anand (Gujarat), India.

(Corresponding author: Prajapati M.D.)*
(Received: 19 April 2025; Revised: 28 May 2025; Accepted: 24 June 2025; Published online: 10 July 2025)
(Published by Research Trend)

ABSTRACT: Root rot caused by *Macrophomina phaseolina* poses a significant threat to black gram (*Vigna mungo*) production in India. This study aimed to isolate and evaluate antagonistic bacterial isolates from the rhizosphere of various trees in Gujarat for their biocontrol potential against *M. phaseolina*. Twelve bacterial isolates were screened in vitro, with five exhibiting over 70% mycelial growth inhibition. Among these, *Pseudomonas aeruginosa* (AF 4) showed the highest antagonistic activity. Seed biopriming with these isolates under pot conditions significantly improved seed germination, seedling vigour and reduced mortality. Biochemical assays revealed enhanced total phenol and peroxidase activities in bacterial-treated plants, peaking at 45 days after sowing, indicating activation of plant defense mechanisms. A significant negative correlation was observed between peroxidase activity and seedling mortality. These findings suggest that antagonistic bacteria, particularly *P. aeruginosa* (AF 4), can serve as effective and eco-friendly biocontrol agents against *M. phaseolina* in black gram.

Keywords: Biopriming, Black gram, *Macrophomina phaseolina*, Antagonistic bacteria, Peroxidase, Total phenol, Disease suppression.

INTRODUCTION

Black gram (*Vigna mungo* L.) is a vital pulse crop in India, valued for its protein content and soil fertility benefits (Gopalan *et al*., 1971). However, its productivity is severely affected by root rot disease caused by *Macrophomina phaseolina*, a pervasive soil- and seed-borne pathogen (Nicholson *et al*., 1972). Conventional chemical control methods have been largely ineffective, necessitating sustainable alternatives (Baird *et al*., 2003).

Plant growth-promoting rhizobacteria (PGPR) have gained prominence as biocontrol agents due to their antagonistic activity against pathogens and their role in enhancing plant resistance (Streit *et al*., 1996; Patten and Glick 2002). Seed biopriming with PGPR facilitates bacterial colonization on seeds, promoting early seedling vigor and protection (Subba Rao and Dommergues 1998). PGPR produce phytohormones such as indole-3-acetic acid (IAA) that enhance root development and nutrient uptake (McQuilken *et al*., 1998). They also induce plant defense responses by increasing phenolic compounds, siderophore production and hydrogen cyanide release, which inhibit pathogen growth (Nicholson and Hammerschmidt 1992; Kuc, 1995).

Moreover, PGPR trigger induced systemic resistance (ISR), characterized by lignin deposition and activation of antioxidant enzymes like peroxidase and superoxide dismutase, strengthening plant defenses (Shoresh *et al*., 2010; Singhai *et al*., 2011). This study investigates native rhizospheric bacteria for their ability to suppress *M. phaseolina* through seed biopriming under controlled conditions, aiming to develop an eco-friendly strategy for managing root rot in black gram (Harborne, 1988).

MATERIAL AND METHODS

The study was conducted at the Department of Plant Pathology, BACA, Anand Agricultural University, Gujarat from 2023 to 2025. Pot experiments were arranged in a completely randomized design (CRD) with six treatments and four replications. The procedure was maintained on specific protocols; these are as follows.

A. Isolation of Rhizospheric Bacteria

Soil samples from rhizospheres of various tree species in Gujarat were collected and processed using the serial

dilution method. Dilutions were plated on Nutrient Agar (NA) and King's B media, incubated at 28 ± 2°C for 48 hours. Distinct colonies were purified by repeated streaking and stored at 4°C.

B Isolation and Identification of M. phaseolina

Black gram roots showing root rot symptoms were collected, surface sterilized with sodium hypochlorite (1%) and plated on Potato Dextrose Agar (PDA). After incubation at 28 ± 1°C for 3–4 days, single hyphal tip isolation was performed to obtain pure cultures. Pathogenicity was confirmed by inoculating healthy soil with fungal cultures followed by sowing black gram seeds. Infected seedlings showed characteristic symptoms and the pathogen was re-isolated, confirming Koch's postulates.

C. In Vitro Antagonistic Screening

Fourteen bacterial isolates and one standard control (*Pseudomonas fluorescens*) were screened using dual culture assay. Mycelial discs of *M. phaseolina* were placed centrally on NA-PDA plates and bacterial suspensions were inoculated 3 cm away. Radial growth of the pathogen was recorded on days 5 and 7 to calculate percent inhibition using Vincent's formula.

D. Morphological, Biochemical and Molecular Characterization

Five effective bacterial isolates—AC 2, AN 3, AF 4, JB 8, NP 9—were characterized in detail. Colony morphology, fluorescence under UV light on King's B medium and Gram staining were documented. Biochemical tests included HCN production, ACC deaminase activity and enzyme production (chitinase, protease, lipase). Antibiotic resistance was assessed via disc diffusion assay. Molecular characterization involved 16S rDNA amplification using U27f/U1492r primers and ARDRA analysis using *Alu*I, *Rsa*I and *Taq*I enzymes to determine genetic diversity.

E. Seed Biopriming and Pot Trials

The treatments applied in this study included biopriming black gram seeds with different bacterial isolates followed by inoculation with *Macrophomina phaseolina.* Treatment 1 (T1) consisted of *Bacillus licheniformis* (AC 2) combined with *M. phaseolina.* Treatment 2 (T2) involved *Bacillus stratosphericus* (AN 3) plus *M. phaseolina.* Treatment 3 (T3) included *Pseudomonas aeruginosa* (AF 4) with *M. phaseolina.* Treatment 4 (T4) was *Pseudomonas azotoformans* (JB 8) plus *M. phaseolina*. Treatment 5 (T5) consisted of *Stenotrophomonas* sp. (NP 9) combined with *M. phaseolina.* Treatment 6 (T6) served as the control, where seeds were inoculated with *M. phaseolina* only, without any bacterial treatment.

Seeds of black gram (*var.* GAU-4) were surface sterilized using 1% sodium hypochlorite to eliminate surface contaminants. For biopriming, seeds were coated with individual bacterial isolates using 1% carboxymethyl cellulose (CMC) as a sticker agent, following the method of Kumar *et al.* (2020). The seeds were then soaked in bacterial suspensions for 5 hours on a shaker to ensure uniform bacterial adhesion and subsequently air-dried at room temperature.

For pathogen inoculation, *Macrophomina phaseolina* was cultured on a sand–maize meal medium (9:1) for 15 days. Sterilized soil was prepared by autoclaving at 121 °C for 5 hours and then inoculated with the fungal-infested sand–maize medium at a rate of 50 g per kg of soil. The inoculated soil was incubated for one week to allow for pathogen establishment.

Bioprimed seeds were sown in earthen pots containing the inoculated soil at a density of five seeds per pot under controlled polyhouse conditions (Fig. 1). Seed germination was recorded 10 days after sowing, followed by further observations at 4 weeks to assess seedling growth parameters such as root and shoot length, vigor index and seedling mortality. The percentage of seed germination and seedling mortality were calculated according to the formula described by Pandey *et al*. (1989). This setup allowed for evaluating the efficacy of bacterial biopriming in enhancing black gram resistance against *M. phaseolina* under pathogen pressure.

Seedling Vigour Index -I= (Mean root length + Mean shoot length) Germination (%)

F. Biochemical Assays

Phenolic compounds are widely recognized for their antimicrobial properties and play a crucial role in enhancing plant defense against a broad spectrum of pathogens, including fungi, bacteria and viruses. In this study, elevated levels of phenolic compounds were observed in the roots, collar region and leaves of black gram plants treated with beneficial bacteria and challenged with pathogenic stress. This increase is likely due to the hydrolysis of phenolic glycosides by fungal glycosidases, releasing free phenols that contribute to restricting pathogen spread (Doughari, 2015).

Similarly, increased peroxidase (PO) activity is strongly correlated with enhanced disease resistance and lignin biosynthesis in plants (Reuveni *et al.*, 1992). At later developmental stages, PO facilitates the cross-linking of hydroxyproline-rich glycoproteins (HRGPs) during lignification, thereby reinforcing cell walls and forming a physical barrier against pathogen invasion. Furthermore, hydrogen peroxide (H_2O_2) generated by PO exhibits potent antimicrobial activity, directly inhibiting pathogens and promoting the generation of reactive oxygen species (ROS) with strong antifungal properties (Chen *et al.*, 2000). Collectively, these biochemical responses underscore the pivotal role of phenolic compounds and PO activity in the induced defense mechanisms of bioprimed black gram plants.

Total phenol content was quantified following the method of Sadasivam and Manickam (1992) using methanolic extracts of plant tissues. Peroxidase activity was measured by the guaiacol-H_2O_2 assay as described by Bergmeyer (1974) with minor modifications. Samples from roots, collar region and leaves were collected at 15, 30 and 45 days after sowing (DAS) from both bioprimed and untreated control plants to assess biochemical changes associated with pathogen resistance.

For total phenol estimation, 200 mg of fresh tissue from each plant part was homogenized in 80% methanol and

the volume was adjusted to 10 ml. After centrifugation at 8000 rpm for 10 minutes, 0.2 ml of the supernatant was used in the assay. The extract was diluted to 1 ml with distilled water, mixed with 1 ml of Folin-Ciocalteau reagent (diluted 1:2) and after 3 minutes, 1 ml of 20% sodium carbonate was added. The mixture was incubated in a boiling water bath for 1 minute, cooled and then diluted to 5 ml with distilled water. Absorbance was measured at 620 nm. Phenol content was calculated using a standard curve and expressed as mg per gram of fresh weight.

Peroxidase activity was determined by homogenizing 0.1 g of fresh tissue in 2 ml potassium phosphate buffer, followed by centrifugation at 16,000 rpm and 4°C for 15 minutes. A reaction mixture containing 0.05 ml of supernatant, 1.5 ml guaiacol and 0.5 ml hydrogen peroxide was prepared and the increase in absorbance due to guaiacol oxidation was recorded at 420 nm at 30-second intervals over 3 minutes. Peroxidase activity was expressed as the change in optical density per minute per gram of fresh tissue (ΔOD/min/g).

G. Statistical Analysis

Data were analyzed using ANOVA (Steel and Torrie 1980). Treatment means were compared using critical difference at 5% level. Correlation between peroxidase activity and seedling mortality was calculated at 45 DAS.

T1- *B. licheniformis* (AC 2) + *M. phaseolina*; T2- *B. stratosphericus* (AN 3) + *M. phaseolina*; T3- *P. aeruginosa* (AF 4) + *M. phaseolina*; T4- *P. azotoformans* (JB 8) + *M. phaseolina*; T5- *Stenotrophomonas* sp. (NP 9) + *M. phaseolina*; T6- Control (*M. phaseolina*)

Fig. 1. Germination of bio primed black gram seeds 10 DAS.

RESULTS AND DISCUSSION

A. Isolation and Screening of Bacterial Isolates

Twelve bacterial isolates were obtained and tested against *M. phaseolina*. Five isolates (AC 2, AN 3, AF 4, JB 8, NP 9) demonstrated over 70% inhibition of fungal growth. AF 4 showed the highest inhibition (74.81%), followed by JB 8 (74.07%) and AC 2 (71.85%). These results are in line with earlier reports where PGPRs like Bacillus and Pseudomonas exhibited antagonism through antibiotic production and lytic enzyme secretion. Related results were observed in Gupta *et al*. (2001).

B. Characterization of Antagonists

AF 4 and JB 8 showed fluorescence under UV light, suggesting pseudomonads. Gram staining classified AC 2 and AN 3 as Gram-positive rods (likely *Bacillus* spp.), while the remaining were Gram-negative rods. Enzymatic assays revealed production of lipase in all isolates, protease in AC 2, AF 4, JB 8, NP 9 and chitinase in AN 3 and JB 8. HCN production was positive for AF 4 and NP 9. ACC deaminase activity, an indicator of stress tolerance, was strong in AF 4 and JB 8. Similar results were obtained in Chandra Nayaka *et al*. (2010).

C. Antibiotic Resistance Profile

All isolates showed resistance to several antibiotics, notably Penicillin-G, Carbenicillin and Ampicillin. AF 4 and JB 8 were susceptible to Streptomycin, Gentamicin and Ciprofloxacin. These patterns are consistent with previous observations of variability in antibiotic resistance among soil bacteria.

D. Molecular Identification and Phylogenetic Analysis

16S rDNA sequencing confirmed identities as *Bacillus licheniformis, B. stratosphericus, P. aeruginosa, P. azotoformans* and *Stenotrophomonas* sp. ARDRA profiles further supported these classifications, revealing high genetic diversity. Phylogenetic clustering showed close relatedness between AC 2 and AN 3 and between AF 4 and JB 8, while NP 9 was genetically distinct.

E. SEM Imaging

Scanning electron microscopy (SEM) of dual culture assay revealed physical damage to *M. phaseolina* hyphae in the presence of *B. licheniformis* (AC 2), including hyphal shrinkage and collapse, confirming mycoparasitic interaction.

F. Effect of Biopriming on Disease Management

AF 4-treated seeds showed 100% germination and maximum seedling vigour (2765), while the control showed only 70% germination and a lower vigour index (1030). As shown in Table 1, Shoot and root lengths were significantly higher in AF 4 (22.45 cm and 5.20 cm, respectively) compared to the control (11.53 cm and 3.18 cm). Mortality was reduced to 20% in AF 4, compared to 60% in untreated controls. Similar effects were observed with JB 8 and AC 2 treatments, indicating broad biocontrol efficacy (Fig. 2). The finding also correlated with the work of Patil *et al*. (2016) who reported that seed treatment with *P. aeruginosa* significantly improved soybean growth under saline conditions, with increases in germination (24.39%), root length (73.97%), shoot length (30.02%) and vigor index (78.97%). It also reduced charcoal rot incidence caused by *M. phaseolina* in both normal and saline soils. Similarly, El-Mougy and Abdel-Kader (2008); Chandra Nayaka *et al*. (2010); Begum *et al*. (2010) found that biopriming seeds of faba bean, maize and soybean enhanced germination, vigor, yield and reduced root rot and damping-off diseases.

Typical rotted seedlings obtained from control pots

T1- *B. licheniformis* (AC 2) + *M. phaseolina*; T2- *B. stratosphericus* (AN 3) + *M. phaseolina*; T3- *P. aeruginosa* (AF 4) + *M. phaseolina*; T4- *P. azotoformans* (JB 8) + *M. phaseolina*; T5- *Stenotrophomonas* sp. (NP 9) + *M. phaseolina*; T6- Control (*M. phaseolina*)

Fig. 2. Effect of seed biopriming with antagonistic bacterial isolates against disease caused by *M. phaseolina* in black gram under pot conditions.

Table 1: Effect of seed biopriming with antagonistic bacterial isolates against root rot of black gram under pot conditions.

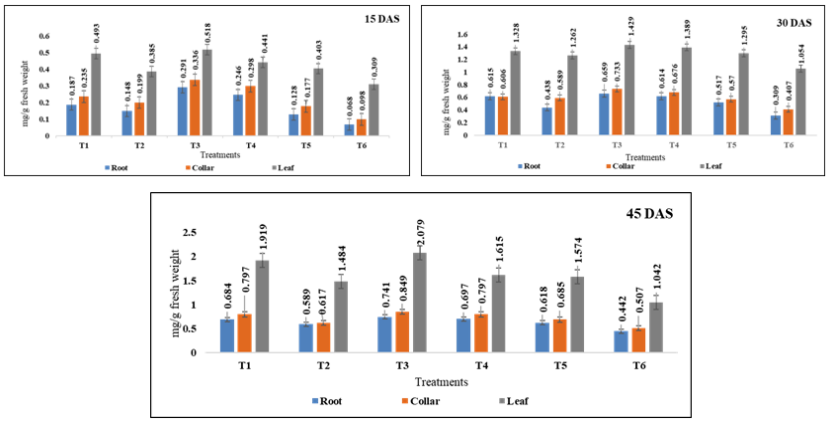
Tr. No.	Treatments	Germination (%)	Shoot length (cm)	Root length (cm)	Seedling Vigour Index-I	Mortality (%)
T_1	*B. licheniformis* (AC 2) + *M. phaseolina*	100	20.25	4.43	2468	26.55 (20.00)
T_2	*B. stratosphericus* (AN 3) + *M. phaseolina*	95	21.38	4.00	2411	39.22 (40.00)
T_3	*P. aeruginosa* (AF 4) + *M. phaseolina*	100	22.45	5.20	2765	26.55 (20.00)
T_4	*P. azotoformans* (JB 8) + *M. phaseolina*	100	21.95	5.18	2713	26.55 (20.00)
T_5	*Stenotrophomonas* sp. (NP 9) + *M. phaseolina*	95	18.70	3.90	2147	29.72 (25.00)
T_6	Control (*M. phaseolina*)	70	11.53	3.18	1030	50.7 (60.00)
S. Em. ±		3.73	0.23	0.11	-	1.29
C. D. at 5%		11.16	0.67	0.32	-	3.84
C. V. (%)		7.99	2.34	4.96	-	7.78

G. Biochemical Induction of Plant Defense

Phenol content increased over time and was highest in leaf tissues at 45 DAS, especially in T3 (Fig. 3). Total phenol content was highest in the leaves, followed by collar and roots, peaking at 45 DAS. The maximum was recorded in AF 4-treated leaves (2.079 mg/g fresh weight) as depicted in Table 2. This suggests a strong defense response, corroborated by Sarma *et al*. (2002), who linked phenolic accumulation to pathogen resistance. The greater accumulation of antifungal phenolic compounds is found above collar regions following infection which may account for the plausible explanation as to why the upper stem portion and leaves are not infected with root rot pathogen (Sarma *et al*., 2002).

Table 2: Phenol accumulation in tissues to pathogen resistance.

Tissue	T3 (AF 4)	Control
Leaf	2.079 mg/g	1.042 mg/g
Collar	0.849 mg/g	0.507 mg/g
Root	0.741 mg/g	0.442 mg/g



T1- *B. licheniformis* (AC 2) + *M. phaseolina*; T2- *B. stratosphericus* (AN 3) + *M. phaseolina*; T3- *P. aeruginosa* (AF 4) + *M. phaseolina*; T4- *P. azotoformans* (JB 8) + *M. phaseolina*; T5- *Stenotrophomonas* sp. (NP 9) + *M. phaseolina*; T6- Control (*M. phaseolina*)

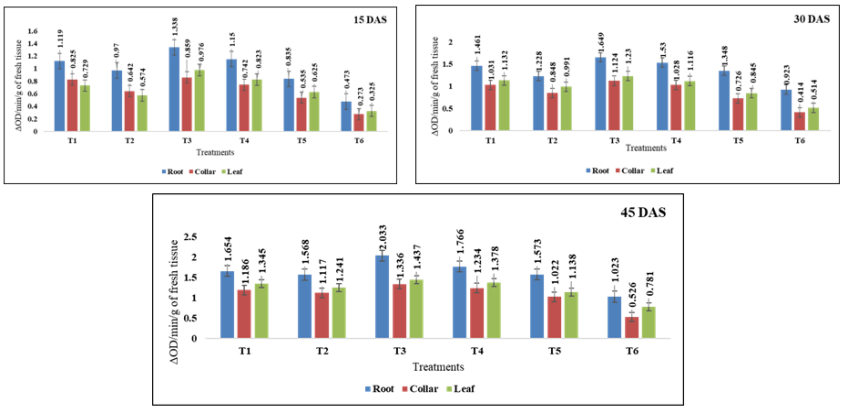
Fig. 3. Phenol content in black gram root, collar and leaf tissue at 15, 30 and 45 DAS

Peroxidase activity was highest in roots and increased over time, peaking at 45 DAS with 2.033 ΔOD/min/g in AF 4-treated plants, as depicted in Table 3. These findings suggest that biopriming stimulates defense-related metabolic responses in black gram. Peroxidase activity is generally higher in roots than in leaves (Fig. 4) due to the roots' primary role in nutrient and water absorption, which exposes them to greater oxidative stress caused by reactive oxygen species (ROS). This oxidative stress triggers the activation of peroxidase enzymes as a protective response.

The observed increase in peroxidase activity at 45 days after sowing (DAS), compared to 15 and 30 DAS, can be attributed to the plant's progression in growth and development, which intensifies the need for ROS-scavenging enzymes like peroxidase to mitigate oxidative damage. Furthermore, the presence of antagonistic bacteria introduced through seed biopriming may have enhanced the plant's innate defense mechanisms, contributing to the elevated peroxidase activity at the later growth stage, as supported by Hasanuzzaman *et al*. (2021).

Table 3: Peroxidase accumulation in tissues to pathogen resistance.

Tissue	T3 (AF 4)	Control
Leaf	1.437 ΔOD/min/g	0.781 ΔOD/min/g
Collar	1.336 ΔOD/min/g	0.526 ΔOD/min/g
Root	2.033 ΔOD/min/g	1.023 ΔOD/min/g



T1- *B. licheniformis* (AC 2) + *M. phaseolina*; T2- *B. stratosphericus* (AN 3) + *M. phaseolina*; T3- *P. aeruginosa* (AF 4) + *M. phaseolina*; T4- *P. azotoformans* (JB 8) + *M. phaseolina*; T5- *Stenotrophomonas* sp. (NP 9) + *M. phaseolina*; T6- Control (*M. phaseolina*)

Fig. 4. Peroxidase activity in black gram root, collar and leaf tissue at 15, 30 and 45 DAS.

H. Correlation Between Mortality and Peroxidase Activity

A highly significant negative correlation (roots: r = -0.856; collar: r = -0.878; leaves: r = -0.877) was observed between peroxidase activity and seedling mortality across all tissues, highlighting the biochemical basis for disease suppression showed in Table 4. Peroxidase is a crucial defense enzyme in plants against diseases caused by pathogens. This confirms the protective role of peroxidase in limiting pathogen spread and enhancing resistance (Fig. 5).

Table 4: Correlation between mortality and peroxidase activity at 45 DAS.

Tissue	Mortality (%)
Root	-0.856**
Collar	-0.878**
Leaf	-0.877**

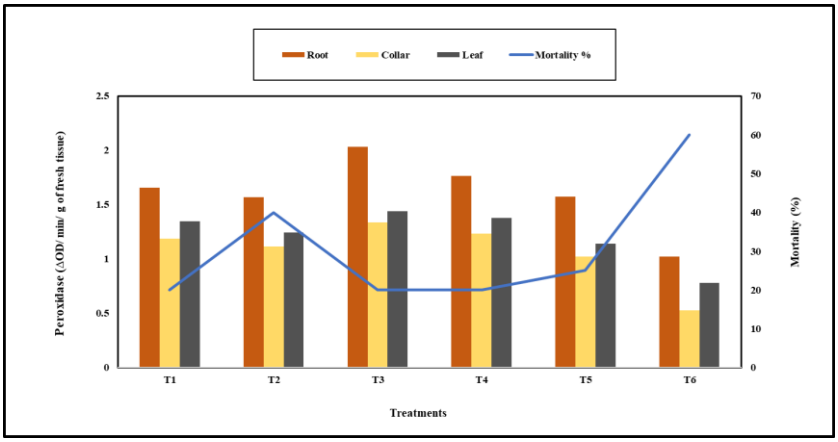


Fig. 5. Correlation between mortality and peroxidase activity of at 45 DAS.

CONCLUSIONS

Seed biopriming with antagonistic bacterial isolates significantly improved plant growth and suppressed root rot caused by *M. phaseolina* in black gram. *Pseudomonas aeruginosa* (AF 4) was the most effective, promoting germination, vigour and biochemical defense activation. This approach offers a promising, sustainable alternative to chemical fungicides.

FUTURE SCOPE

Further research is needed to evaluate the efficacy of these isolates under field conditions and diverse agro-climatic zones. Development of stable bioinoculant formulations, elucidation of molecular pathways involved in induced resistance and integration with other biocontrol agents may enhance practical applications.

Acknowledgement. The author thanks the Department of Plant Pathology, Anand Agricultural University, for providing the necessary facilities and guidance. Special appreciation to advisory committee members and technical staff for their support.

Conflict of Interest. None.

REFERENCES

Begum, M., Sariah, M., Puteh, A., Zainal Abidin, M., Rahman M. and Siddiqui, Y. (2010). Field performance of bio-primed seeds to suppress *Colletotrichum truncatum* causing damping-off and seedling stand of soybean. *Biological Control*, *53*, 18-23.

Bergmeyer, H. U. (1974). Peroxidase, Methods of Enzymatic Analysis, *2*, 685-690.

Baird, R. E., Watson, C. E. and Scruggs, M. (2003). Relative longevity of *Macrophomina phaseolina* and associated mycobiota on residual soybean roots in soil. *Plant Disease*, *87*(5), 563-566.

Chandra Nayaka, S., Niranjana, S. R., Uday Shankar, A. C., Niranjan Raj, S., Reddy, M. S., Prakash, H. S. and Mortensen, C. N. (2010). Seed biopriming with novel strain of Trichoderma harzianum for the control of toxigenic Fusarium verticillioides and fumonisins in maize. *Archives of Phytopathology and Plant Protection*, *43*(3), 264-282.

Chen, C., Belanger, R. R., Benhamou, N. and Paulitz, T. C. (2000). Defense enzymes induced in cucumber roots by treatment with plant growth-promoting rhizobacteria (PGPR) and *Pythium aphanidermatum*. *Physiological and Molecular Plant Pathology*, *56*(1), 13-23.

Doughari, J. (2015). An overview of plant immunity. *Journal of Plant Pathology and Microbiology*, *6* (11), 1-11.

El-Mougy, N. and Abdel-Kader, M. (2008). Long term activity of biopriming seed treatment for biological control of faba bean root rot pathogens. *Australian Plant Pathology*, *37*, 464-471.

Gupta, C. P., Dubey, R. C., Kang, S. C. and Maheshwari, D. K. (2001). Antibiosis- mediated necrotrophic effect of Pseudomonas GRC 2 against two fungal plant pathogens. *Current Science*, *81*(1), 91-94.

Gopalan, C., Shastri, B. V. and Balasubramaniam, S. C. (1971). Nutritive value of Indian foods. National Institute of Nutrition, Indian Council of Medical Research (ICMR), Hyderabad.

Hasanuzzaman, M., Parvin, K., Bardhan, K., Nahar, K., Anee, T. I., Masud, A. A. C. and Fotopoulos, V. (2021). Biostimulants for the regulation of reactive oxygen species metabolism in plants under abiotic stress. *Cells*, *10*(10), 25-37.

Harborne, J. B. (1988). Introduction to ecological biochemistry, 3rd edition. Academic Press, London.

Kuc, J. (1995). Induced systemic resistance-an overview. Induced Resistance to Disease in Plants, 169-175.

Kumar, P., Aeron, A., Shaw, N., Singh, A., Bajpai, V. K., Pant, S. and Dubey, R. C. (2020). Seed bio-priming with tri-species consortia of phosphate solubilizing rhizobacteria (PSR) and its effect on plant growth promotion. *Heliyon*, *6*(12).

Nicholson, J. F., Dhingra, O. D. and Sinclair, J. B. (1972). Internal seed-borne nature of *Sclerotinia sclerotiorum* and *Phomopsis* sp. *Phytopathology*, *62*, 1261- 1263.

Nicholson, R. L. and Hammerschmidt, R. (1992). Phenolic compounds and their role in disease resistance. *Annual Review of Phytopathology*, *30*(1), 369-389.

McQuilken, M. P., Halmer, P. and Rhodes, D. J. (1998). Application of microorganisms to seeds. *Formulation of Microbial Biopesticides: Beneficial Microorganisms, Nematodes and Seed Treatments*, 255-285.

Pandey, H., Shukla, K. and Tewari, G. (1989). Effect of moisture stress, plant population density and pathogen inoculation on charcoal stalk rot of sorghum, *Annals of Applied Biology*, *116*, 221-232.

Patten, C. L. and Glick, B. R. (2002). Role of Pseudomonas putida indole acetic acid in development of the host plant root system. *Applied and Environmental Microbiology*, *68*(8), 3795-3801.

Patil, S., Paradeshi, J. and Chaudhari, B. (2016). Suppression of charcoal rot in soybean by moderately halotolerant *Pseudomonas aeruginosa* GS-33 under saline

conditions. *Journal of Basic Microbiology*, *56*(8), 889-899.

Reuveni, R., Shimoni, M., Karchi, Z. and Kuc, J. (1992). Peroxidase activity as a biochemical marker for resistance of muskmelon (*Cucumis melo*) to *Pseudoperonospora cubensis*. *Phytopathology*, *82*(7), 749-753.

Sadasivam, S. and Manickam, A. (1992). Biochemical methods for agricultural sciences, *Wiley Eastern Limited, New Delhi, pp.* 11-12.

Sarma, B. K., Singh, D. P., Mehta, S., Singh, H. B. and Singh, U. P. (2002). Plant growth- promoting rhizobacteria-elicited alterations in phenolic profile of chickpea (*Cicer arietinum*) infected by *Sclerotium rolfsii*. *Journal of Phytopathology*, *150*(4), 277-282.

Shoresh, M., Harman, G. E. and Mastouri, F. (2010). Induced systemic resistance and plant responses to fungal biocontrol agents. *Annual Review of Phytopathology*, *48*, 21-43.

Singhai, P. K., Sarma, B. K. and Srivastava, J. S. (2011). Biological management of common scab of potato through *Pseudomonas* spp. and vermicompost. *Biological Control*, *57*(2), 150-157.

Steel, R. G. and Torrie, J. H. (1980). Principles and procedures of statistics: a biometrical approach. New York: *McGraw-Hill, 2*, 137-139.

Streit, W. R., Joseph, C. M. and Phillips, D. A. (1996). Biotin and other water-soluble vitamins are key growth factors for alfalfa root colonization by *Rhizobium meliloti* 1021. *Molecular Plant-Microbe Interactions: MPMI*, *9*(5), 330-338.

Subba Rao, N. S. and Dommergues, Y. R. (1998). *Microbial Interactions in Agriculture and Forestry.*

How to cite this article: Prajapati M.D., Patel H.H., Kandoriya P.J., Joshi R.L. and Chaudhari A.K. (2025). Antipathogenic Activity of Antagonistic Bacterial Isolates Against *Macrophomina phaseolina* and their Role in Inducing Biochemical Defence in Black Gram under Pot Conditions. *Biological Forum, 17*(7): 102-108.