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Integrated biocontrol of okra root rot caused by *Fusarium solani* and *Macrophomina phaseolina* using *Pseudomonas fluorescens* and *Saccharomyces cerevisiae*

Ahmed Falih Shamukh
Plant Protection Department, College of Agriculture, University of Misan, Iraq. E-mail: ahmed.faleh@uomisan.edu.iq

Norien Abdulzahra Hasan
Plant Protection Department, College of Agriculture, University of Misan, Iraq. E-mail: norienabulzahra@uomisan.edu.iq

Wedad Marid Hamood
Plant Protection Department, College of Agriculture, University of Misan, Iraq. E-mail: wedadmarid@uomisan.edu.iq

Qusai Hattab Madhi
* Corresponding author. Plant Protection Department, College of Agriculture, University of Misan, Iraq. E-mail: qusay.hattab@uomisan.edu.iq

Abstract

Objective

This study evaluated the antagonistic potential of the yeast *Saccharomyces cerevisiae* and the bacterium *Pseudomonas fluorescens* against *Fusarium solani* and *Macrophomina phaseolina*, the causal agents of okra (*Abelmoschus esculentus* L.) root rot and seedling damping-off. The research involved isolation and identification of the pathogens, assessment of their pathogenic variability, and evaluation of biological control agents under laboratory and greenhouse conditions.

Materials and methods

Three isolates of *F. solani* and two isolates of *M. phaseolina* were obtained from infected okra plants collected from different locations. Significant variation in pathogenicity was observed among the isolates. The isolate F_1 of *F. solani* exhibited the highest virulence, causing a significant reduction in seed germination and increasing seedling mortality by 43.2% and 49.4%, respectively, compared with the untreated control.

Results

In vitro assays demonstrated that *P. fluorescens* effectively inhibited the radial growth of *F. solani* and *M. phaseolina* by 48.36% and 64.7%, respectively. Similarly, *S. cerevisiae* exhibited significant antifungal activity, particularly at the fifth dilution, where growth inhibition reached

74.1% for *F. solani* and 76.1% for *M. phaseolina*. These findings demonstrate that both biological agents significantly suppressed fungal growth under in vitro conditions. Pathogenicity tests confirmed that infection with *F. solani* and *M. phaseolina* significantly reduced seed germination and increased seedling mortality. However, application of biological treatments reduced disease severity. Among all treatments, the combined application of *M. phaseolina* with *P. fluorescens* produced the best results, significantly improving germination percentage and reducing seedling mortality compared to pathogen-only treatments. Greenhouse experiments further revealed that both *S. cerevisiae* and *P. fluorescens*, applied individually or in combination, significantly enhanced seed germination, chlorophyll content, vegetative growth, and leaf area. Moreover, the combined treatment exhibited a strong synergistic effect in suppressing pathogen activity, reducing disease incidence and severity, and promoting overall plant growth performance.

Conclusion

Overall, the results of this study demonstrate that *S. cerevisiae* and *P. fluorescens* are promising eco-friendly biological control agents that can be effectively used in integrated management strategies for controlling okra root rot disease caused by *F. solani* and *M. phaseolina*. These findings support the development of sustainable disease management approaches that reduce reliance on chemical fungicides while improving plant health and productivity.

Keywords: *Biocontrol agents, Fusarium solani, Macrophomina phaseolina, Pseudomonas fluorescens, Saccharomyces cerevisiae*

Paper Type: Research Paper.

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Introduction

Okra (*Abelmoschus esculentus* L.) belongs to the Malvaceae family and is cultivated in tropical, subtropical, warm, and temperate regions (Ounis et al., 2024). Global okra production reached 11.23 million tons from 2.80 million hectares, an increase of 5.90% over the previous year (FAOSTAT, 2022). Okra is an important vegetable crop with high nutritional value and numerous health benefits. It can be consumed fresh, and its fruits (the gum, seeds, and pods) contain bioactive compounds. The phytochemicals in okra demonstrate therapeutic activity

against several chronic diseases, such as type 2 diabetes, gastrointestinal disorders, and cardiovascular diseases (Elkhalifa et al., 2021; Meiriani & Tarigan, 2024). In Iraq, okra is widely cultivated for fresh consumption, freezing, and drying, especially during the winter season. Okra pods play a vital role in meeting nutritional needs (Meiriani & Tarigan, 2024). They are rich in calcium and contribute significantly to the human diet by providing fats, proteins, carbohydrates, minerals, and vitamins such as riboflavin (B2), thiamin (B1), vitamin C, and vitamin A. Mature seeds contain approximately 20% edible oil (Benchasri, 2012). Okra is susceptible to several fungal diseases, including seed rot, damping-off, and Fusarium wilt, which significantly impact both the quantity and quality of the crop (Appiah et al., 2020). Farmers often rely on chemical pesticides; however, these pesticides cause problems such as pest resistance, adverse effects on animals, pesticide residues on plants, and environmental pollution (USEPA, 2023). Biological control has emerged as a promising alternative due to its ability to suppress pathogens through mechanisms such as competition for nutrients, antibiotic production, and stimulation of plant defense responses. Yeast contains natural plant hormones and hormone-like compounds such as auxins, cytokinins, and gibberellins, which stimulate cell division and expansion, protein and nucleic acid synthesis, chlorophyll formation, and delay leaf senescence (Abd-Elbaky et al., 2021; Abd-Alrahman & Aboud, 2021). Yeast extracts are rich in proteins, minerals, carbohydrates, reducing sugars, lipids, enzymes, nucleic acids, folic acid, and vitamins, including B complex vitamins (Al-Juthery et al., 2020). According to Mankoti et al. (2024) and Taylor et al. (2024), *P. fluorescens* plays a crucial role in promoting plant growth and biological control, as its bioactive molecules demonstrate strong potential in plant growth and defense against pathogens. Root rot caused by *Fusarium solani* and *Macrophomina phaseolina* is among the most destructive soil-borne diseases of okra, leading to poor seed germination, damping-off, reduced plant vigor, and substantial yield losses, particularly under favorable environmental conditions. Although chemical fungicides are commonly used to manage these pathogens, their repeated application may result in environmental contamination, pathogen resistance, and negative effects on non-target organisms. Consequently, environmentally friendly biological control agents such as *Pseudomonas fluorescens* and *Saccharomyces cerevisiae* have attracted considerable attention because of their ability to suppress soil-borne pathogens through multiple mechanisms while promoting plant growth and supporting sustainable crop production. Therefore, the objectives of this study were to isolate and identify the causal agents of okra root rot, evaluate their pathogenicity, and investigate the effectiveness of *Pseudomonas fluorescens* and *Saccharomyces cerevisiae* as biological control agents under laboratory and greenhouse conditions.

Materials and methods

Isolation and identification of fungi from infected okra roots: Samples of okra plants exhibiting root rot were collected from the Al-Batira area in Misan Governorate. Root samples were washed thoroughly under running tap water for 30 min, air-dried on Whatman No. 4, and then cut into small pieces (0.5-1 cm). The pieces were surface-sterilized in 10% sodium hypochlorite solution for 2-3 minutes, rinsed with sterile distilled water, dried, and placed on aqueous agar medium (20 g agar/L + 250 mg chloramphenicol/L). Plates were incubated at 25 ±

2° C for seven days, after which growing fungal colonies were transferred to PSA medium for purification. Identification to species level was based on standard taxonomic keys (Nelson et al., 1983; Summerell et al., 2003).

Pathogenicity experiment: Pathogenicity experiment of the isolated fungi was examined on water agar plates. A 0.5 cm disc from a 7-day-old colony was placed at the center of each plate, with three replicates for each isolate. Sterile okra seeds (surface-sterilized with 2% sodium hypochlorite) were placed in a circular pattern around the colony edge (20 seeds per plate). Plates were incubated at 25 ± 2° C, and germination percentage was recorded after 72 hours. Healthy and diseased seedlings were counted after two weeks. According to the following formula:

$$\text{Germination\%} = \frac{\text{Number of germinated seeds}}{\text{Total number of seeds}} =\times\ 100$$

Preparation of fungal inoculum: Inoculum of *F. solani* and *M. phaseolina* was prepared on millet seeds (*Panicum milliaceum*) following Smiley (2019).

Evaluation of *Pseudomonas fluorescens* in fungal growth inhibition: The bacterial isolate was obtained from Dr. Diaa Al-Waeli (College of Agriculture, University of Basrah). PDA medium was autoclaved, cooled, and amended with *P. fluorescens* at 1 mL/L. The amended medium was poured into sterile Petri dishes and incubated at 30° C for 24 hours. Each plate was inoculated at the center with a 5 mm fungal disc of *F. solani* or *M. phaseolina*. Control plates lacked bacteria. Plates were incubated at 27° C for 7 days. Radial growth was measured, and inhibition percentage was calculated using Abbott's formula (1925).

Yeast source and activation: Commercial dry baker's yeast (*Saccharomyces cerevisiae*, Altunkaya Co., Turkey) was used as a biocontrol agent. Five grams of yeast were dissolved in 1 L of sterile warm water (30-35° C) with 2 g sucrose. The mixture was incubated at 25° C for 24 hours. The suspension was filtered through Whatman No. 1 and then through a 0.22 µm membrane filter to obtain a cell-free filtrate (Chalutz et al., 1977).

Evaluation of yeast against fungal pathogens: Serial dilutions (10^{-4} and 10^{-5}) of the yeast filtrate were incorporated into PDA medium and poured into sterile Petri dishes. After the medium solidified, each plate was inoculated with a 0.5-cm mycelial disc taken from the actively growing margin of a 7-day-old fungal culture and incubated at 27 ± 1°C. Radial fungal growth was measured daily until the control plates were completely covered by fungal growth. The percentage of growth inhibition was then calculated using the previously described formula.

Field experiment

Evaluation of yeast and bacteria in controlling root rot: A field experiment was conducted at the Experimental Farm, College of Agriculture, University of Misan. The soil was plowed and leveled, and 36 experimental units (2 × 2 m) were prepared under a completely randomized design with 12 treatments and 3 replicates. Seeds were surface-sterilized (1% NaOCl) and planted at 25 cm spacing. Fungal inoculum (2 g per kg of soil) was applied, while bacterial inoculation involved soaking seeds in *P. fluorescens* suspension (~10^8 CFU/mL + 1% Arabic

gum). Yeast suspension (*S. cerevisiae*, 10^7-10^8 cells/mL) was applied at 5 mL per hill. Germination rates seedling mortality and pathogen rei-solation were recorded.

Chlorophyll estimation: Fresh leaves (2 g) were extracted with acetone, and absorbance was measured at 645 and 663 nm. Total chlorophyll was calculated as below (Arnon, 1949):

Total Chlorophyll (mg/L) = 20.2 × A645 + 8.02 × A663

Then, converted to mg/g.

Plant height, stem diameter, and leaf area: The plant height (cm) was measured for ten plants per treatment. The stem diameter was measured using Venire calipers. The leaf area was calculated based on the weight of the fresh leaf and the weight of a 1 cm² leaf disc using the following formula:

Leaf area (cm²) = Leaf weight (g) × Disc area (1 cm²) / Disc weight (g)

Statistical analysis: A completely randomized design (CRD) was applied for laboratory and pot experiments. To compare the means, the least significant difference (LSD) test was applied with a probability 0.05. Statistical program (SPSS) version 23 was used for analyzing data.

Results and discussion

Isolation and identification: Isolation results from infected okra roots revealed three isolates belonging to *F. solani* and two belonging to *M. phaseolina*, indicating the involvement of both fungi in causing the disease symptoms characterized by root rot, wilt, and stunted growth. diseases due to its aggressive colonization of root tissues, disruption of water and nutrient transport, and subsequent physiological deterioration of infected plants (Al-Ghazali et al., 2025; Summerell et al., 2003). The high frequency of this pathogen may also be attributed to its adaptability to diverse soil environments and its ability to survive and spread through infected plant debris and soil. In contrast, *M. phaseolina* is an important soil-borne pathogen responsible for root rot and sudden wilting, particularly under conditions of high temperature and moisture stress. Its capacity to produce microsclerotia enables long-term survival in soil and enhances disease development and severity (El-Rayes et al., 2022; Marquez et al., 2021). Similar findings were reported by Reznikov et al. (2019), who emphasized the strong pathogenic potential of *M. phaseolina* and its significant impact on plant vigor, growth, and stand establishment.

Pathogenicity experiment of *Fusarium solani* and *Macrophomina phaseolina* using okra seeds: Pathogenicity experiment of the three *F. solani* isolates and the two *M. phaseolina* isolates on okra seed germination and seedling mortality (Table 1) revealed that isolate F_1 was the most aggressive, reducing seed germination and increasing seedling mortality to 43.2% and 49.4%, respectively. This was followed by isolates F_2 and F_3, which caused germination and mortality percentages of (55.5, 58.8%) and (60.2, 62.9%), respectively. The *M. phaseolina* isolates Mf_1 and Mf_2 recorded (47.2, 52.3%) and (56.4, 66.4%), respectively, while the germination percentage in the control treatment reached 100%. The ability of *F. solani* to reduce seed germination may be attributed to its production of pectinolytic and cellulolytic enzymes, phytotoxic metabolites, and other virulence factors that damage seed tissues and interfere with normal germination processes. Similar effects were reported by Abiala et al. (2021), who observed significant reductions in seed

germination due to fungal infection. The pathogenicity of *F. solani* is further associated with its capacity to produce cell wall-degrading enzymes and toxins that facilitate host penetration, colonization, and tissue maceration while suppressing plant defense mechanisms (Summerell et al., 2003; Islam et al., 2012). In addition, members of the Fusarium complex possess a wide range of pathogenicity-related genes involved in host invasion, nutrient acquisition, and toxin biosynthesis, contributing to disease development and seedling mortality (Dean et al., 2012; Marquez et al., 2021). These mechanisms ultimately lead to cellular disruption, reduced germination, poor seedling establishment, and severe disease symptoms (Reznikov et al., 2019).

Table 1. Effect of fungal isolates on seed germination and seedling mortality (%)

Isolate	Germination %	Seedling mortality %
F_1	43.2^{d}	49.4^{d}
F_2	55.5^{b}	58.8^{c}
F_3	60.2^{a}	62.9^{b}
M.ph$_1$	47.2^{c}	52.3^{d}
M.ph$_2$	56.4^{b}	66.4^{a}
L.S.D (0.05)	2.4	2.5

Note: Means followed by different letters within each column are significantly different at $P \leq 0.05$ according to LSD test.

Effect of *Pseudomonas fluorescens* on the radial growth of *Fusarium solani* and *Macrophomina phaseolina:* The antagonism test showed that *P. fluorescens* exhibited a clear inhibitory effect on the radial growth of *F. solani* and *M. phaseolina* with inhibition rates of 48.36 and 64.7% respectively. This pronounced antifungal activity may be attributed to the production of a wide range of antimicrobial metabolites, including phenazines, pyoluteorin, and hydrogen cyanide, as well as the secretion of extracellular lytic enzymes that degrade fungal cell walls and restrict mycelial expansion (Nagarajkumar et al., 2004; Haas & Défago, 2005). In addition, *P. fluorescens* is well documented for its ability to compete for nutrients and ecological niches in the rhizosphere and to induce systemic resistance in host plants, thereby enhancing disease suppression beyond direct antagonism (Weller, 2007; Lahlali et al., 2022). These mechanisms collectively explain its strong biocontrol efficacy against soil-borne pathogens such as *Fusarium spp*. and *M. phaseolina*.

Antifungal activity of *Saccharomyces cerevisiae* against *Fusarium solani* and *Macrophomina phaseolina:* The results demonstrated that *S. cerevisiae* significantly inhibited the growth of both tested fungi at the fifth dilution (1:10000) level, with inhibition percentages reaching 74.1% and 76.1%, respectively, compared with the control treatment. This strong antifungal activity can be attributed to multiple antagonistic mechanisms, including competition for nutrients and space, secretion of hydrolytic enzymes, and production of volatile organic compounds (VOCs) with antifungal activity that disrupt fungal growth and sporulation. These findings are consistent with Meng *et al.* (2025), who reported that S. cerevisiae suppresses phytopathogenic fungi through the production of antifungal VOCs, confirming its role as an effective biocontrol agent. Further support is provided by El-Sayed (2022), who demonstrated the

effectiveness of yeast-based biocontrol in reducing soil-borne fungal pathogens. In addition, recent studies have highlighted that antagonistic yeasts exert broad-spectrum antifungal effects against *Fusarium spp.* and *Rhizoctonia solani* through multiple modes of action including enzyme secretion and induction of host resistance (Podgórska-Kryszczuk et al., 2022; Lahlali et al., 2022).

Effect of *Pseudomonas fluorescens* and *S. cerevisiae* and their interaction on okra seed germination and seedling mortality: The results (Table 2) showed that the pathogenic fungi *F. solani* and *M. phaseolina* significantly reduced seed germination and increased seedling mortality to (34.33, 25.13%) and (24.66, 21.21%) respectively compared with the control (100%, 0%). The application of *P. fluorescens* and *S. cerevisiae* individually or combined increased germination and reduced seedling mortality. The best treatment was M.f + P.f, achieving (65.00%, 14.267%) compared with the pathogen-only treatment M.f (34.33%, 13.20%). The reduction in seed germination and increased seedling mortality caused by soil-borne pathogens such as *Fusarium solani* and *M. phaseolina* can be attributed to their aggressive infection strategies, including the production of cell wall-degrading enzymes and phytotoxic metabolites that impair seed metabolism and inhibit normal germination processes. These pathogens are well recognized for their ability to colonize root tissues, disrupt vascular functions, and cause severe physiological dysfunction in infected plants (Ma et al., 2013). The improvement in seed germination and reduction in disease severity following the application of *P. fluorescens* and *S. cerevisiae* can be explained by their multifaceted biocontrol mechanisms.

Table 2. Effect of *Pseudomonas fluorescens* and the yeast *Saccharomyces cerevisiae*, and their interaction, on the germination percentage of okra seeds and seedling mortality in the greenhouse

Treatment	% Germination	% Seedling Mortality
Control	100.00^{a}	0.00^{g}
Pathogen M.f	24.66^{e}	21.21^{b}
Pathogen F.s	34.33^{de}	13.20^{cd}
M.f + Y	53.33^{bc}	25.13^{a}
M.f + P.f	65.00^{b}	14.27^{c}
F.s + Y	55.66^{bc}	17.03^{bc}
F.s + P.f	52.66^{c}	14.86^{c}
M.f + F.s + Y	35.66^{de}	15.13^{c}
F.s + P.f + M.f	51.66^{c}	12.10^{cd}
M.f + F.s + Y + P.f	38.00^{d}	8.16d^{e}
L.S.D (0.05)	11.45	3.23

Note: Means followed by different letters within each column are significantly different at $P \leq 0.05$ according to LSD test. Y = *Saccharomyces cerevisiae*; P.f = *Pseudomonas fluorescens*; F.s = *Fusarium solani*; M.f = *Macrophomina phaseolina*.

These include competition for nutrients and ecological niches, secretion of antimicrobial compounds, and induction of systemic resistance in host plants, which collectively reduce pathogen establishment and disease development *P. Fluorescent* are particularly effective due to their ability to colonize the rhizosphere and produce a wide range of secondary metabolites that suppress fungal growth (Lugtenberg & Kamilova, 2009; Haas & Défago, 2005). Similarly,

antagonistic yeasts exert biocontrol activity through nutrient competition, production of volatile organic compounds, and secretion of hydrolytic enzymes that inhibit fungal development (Spadaro & Droby, 2016). The synergistic effect observed when combining biocontrol agents is consistent with previous studies indicating that integrated microbial applications enhance disease suppression more effectively than single treatments. This enhanced performance is attributed to complementary modes of action, leading to improved plant vigor and reduced pathogen pressure in the rhizosphere (Lahlali et al., 2022).

Effect of *S. cerevisiae* and *P. fluorescens* and their interaction on growth and yield indices of okra: The results presented in Table 3 showed that inoculation with *Fusarium solani* or *Macrophomina phaseolina* significantly reduced leaf area, stem height, stem diameter, and chlorophyll content compared with the healthy control. In contrast, application of *Pseudomonas fluorescens* and/or *Saccharomyces cerevisiae* alleviated the adverse effects of pathogen infection and, in several treatments, significantly improved plant growth parameters beyond those of the untreated control. Leaf area decreased from 6 cm^2 in the control treatment to 3.66 and 1.33 cm^2 for *F. solani* and *M. phaseolina* respectively, with significant differences. Similarly, stem length decreased from 36 cm in the control to 23.66 and 25.00 cm respectively. Chlorophyll content also decreased from 19.33 mg/g to 9.00 and 10.16 mg/g respectively. The reduction in plant growth parameters caused by *F. solani* and *M. phaseolina* is mainly attributed to their ability to colonize root tissues, disrupt vascular transport, and produce cell wall-degrading enzymes and phytotoxic metabolites, which collectively impair water and nutrient uptake and reduce photosynthetic efficiency (Dean et al., 2012; Marquez et al., 2021). In addition, *M. phaseolina* is particularly destructive due to its ability to form microsclerotia, enabling long-term survival in soil and contributing to severe disease development under stress conditions (Reznikov et al., 2019). In contrast, the application of *P. fluorescens* and *S. cerevisiae* significantly improved plant growth and alleviated pathogen-induced damage. This positive effect can be attributed to multiple mechanisms including nutrient solubilization, production of antimicrobial compounds, competition for ecological niches, and induction of systemic resistance in plants. Beneficial rhizobacteria such as Pseudomonas spp. are well known for producing a wide range of antibiotics and secondary metabolites that suppress soil-borne pathogens and promote plant growth (Raaijmakers & Mazzola, 2012). Similarly, biological control agents enhance plant health through rhizosphere colonization and stimulation of plant defense responses, leading to improved growth under pathogen pressure. The combined application of bacterial and yeast biocontrol agents showed enhanced effectiveness compared with individual treatments, suggesting synergistic interactions between different modes of action. Such integrated biological control strategies are considered more stable and effective in suppressing soil-borne diseases, as they combine antibiosis, competition, and induced resistance mechanisms, resulting in improved plant performance under pathogen stress (Ongena & Jacques, 2008; Lahlali et al., 2022). Interestingly, some biocontrol treatments resulted in growth parameters that exceeded those of the healthy control, suggesting that *P. fluorescens* and *S. cerevisiae* not only suppressed pathogen infection but also promoted plant growth through plant growth-promoting mechanisms.

Table 3. Effect of the yeast *Saccharomyces cerevisiae*, the bacterium *Pseudomonas fluorescens*, and their interaction on some growth and yield parameters of okra plants.

Treatment	Leaf Area (cm^2)	Stem Height (cm)	Stem Diameter (cm)	Chlorophyll Content (mg/g)
Control	6.000^{cd}	36.000^{de}	7.000^{abc}	19.33^{de}
M.f	1.333^{f}	25.000^{f}	6.333^{c}	10.16^{f}
F.s	3.667^{e}	23.667^{f}	7.000^{abc}	9.00^{f}
P.f	6.333^{cd}	43.333^{abc}	7.000^{abc}	20.03^{de}
Y	8.000^{abc}	45.000^{ab}	7.000^{abc}	23.04^{cd}
Y + F.s	5.000^{de}	46.000^{a}	7.000^{abc}	13.66^{ef}
Y + M.f	6.000^{cd}	46.333^{a}	6.667^{bc}	13.43^{ef}
P.f + M.f	8.000^{abc}	33.667^{e}	6.667^{bc}	14.46^{ef}
P.f + F.s	6.667^{bc}	42.333^{bcd}	7.667^{ab}	29.39^{b}
Y + F.s + M.f	6.000^{cd}	36.333^{de}	7.667^{ab}	27.16^{bc}
P.f + F.s + M.f	7.000^{abc}	41.667^{bcd}	8.667^{a}	33.60^{a}
P.f + Y + M.p + F.s	8.667^{a}	35.667^{de}	6.667^{bc}	18.87^{de}
L.S.D (0.05)	2.065	9.157	2.159	9.46

Note: Means followed by different letters within each column are significantly different at $P \leq 0.05$ according to LSD test. Y = *Saccharomyces cerevisiae*; P.f = *Pseudomonas fluorescens*; F.s = *Fusarium solani*; M.f = *Macrophomina phaseolina*.

Conclusions: This study demonstrated that *Fusarium solani* and *Macrophomina phaseolina* significantly reduced seed germination and increased seedling mortality in okra. In contrast, *Pseudomonas fluorescens* and *Saccharomyces cerevisiae* as bioagents markedly enhanced seed germination and lowered seedling mortality caused by fungal pathogens. The combined application of these two bioagents exhibited a synergistic effect, effectively suppressing fungal growth and improving overall seedling health. Herein, the application of *P. fluorescens* and S. cerevisiae in biological control program is therefore recommended, along with field evaluations under diverse environmental conditions to validate their efficacy. Further investigations into their antifungal metabolites are also encouraged to support the development of improved biocontrol strategies.

Author contributions

Ahmed Falih Shamukh contributed to the study by proposing the research plan and participating in manuscript writing. Norien Abdulzahra Hasan and Wedad Marid Hamood conducted the experimental work. Qusai Hattab Madhi participated in conducting the experiments, contributed to manuscript writing, and performed the statistical analysis.

Data availability statement

All data generated or analyzed during this study are included in this published article.

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Ethical considerations

This study did not involve human participants or animal experiments. Therefore, ethical approval was not required.the completion of this research.

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Conflict of interest

The authors declare that they have no conflict of interest.

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کنترل زیستی تلفیقی پوسیدگی ریشه بامیه ناشی از *Fusarium solani* و *Macrophomina phaseolina* با استفاده از *Pseudomonas fluorescens* و *Saccharomyces cerevisiae*

احمد فالح شموخ

گروه حفاظت گیاه، دانشکده کشاورزی، دانشگاه میسان، عراق. ایمیل: ahmed.faleh@uomisan.edu.iq

نورین عبد الزهره حسن

گروه حفاظت گیاه، دانشکده کشاورزی، دانشگاه میسان، عراق. ایمیل: norienabulzahra@uomisan.edu.iq

وداد مرید حمود

گروه حفاظت گیاه، دانشکده کشاورزی، دانشگاه میسان، عراق. ایمیل: wedadmarid@uomisan.edu.iq

قصی حطاب ماضی

* نویسنده مسئول. گروه حفاظت گیاه، دانشکده کشاورزی، دانشگاه میسان، عراق. ایمیل: qusay.hattab@uomisan.edu.iq

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چکیده

هدف: این مطالعه با هدف ارزیابی توان آنتاگونیستی مخمر *Saccharomyces cerevisiae* و باکتری *Pseudomonas fluorescens* علیه قارچ‌های *Fusarium solani* و *Macrophomina phaseolina*، عوامل ایجادکننده پوسیدگی ریشه و مرگ گیاهچه (Damping-off) در بامیه (*Abelmoschus esculentus* L.) انجام شد. این پژوهش شامل جداسازی و شناسایی عوامل بیماری‌زا، بررسی تفاوت‌های بیماری‌زایی آن‌ها و ارزیابی عوامل کنترل زیستی در شرایط آزمایشگاهی و گلخانه‌ای بود.

مواد و روش‌ها: سه جدایه از *Fusarium solani* و دو جدایه از *Macrophomina phaseolina* از گیاهان بامیه آلوده جمع‌آوری‌شده از مناطق مختلف جداسازی شدند. بین جدایه‌ها از نظر شدت بیماری‌زایی اختلاف قابل توجهی مشاهده شد. جدایه F1 از *F. solani* بیشترین حدت بیماری‌زایی را نشان داد و در مقایسه با تیمار شاهد، موجب کاهش معنی‌دار درصد جوانه‌زنی بذر و افزایش مرگ‌ومیر گیاهچه‌ها به ترتیب به میزان ۴۳/۲ درصد و ۴۹/۴ درصد شد.

نتایج: آزمون‌های آزمایشگاهی (In vitro) نشان داد که *Pseudomonas fluorescens* رشد شعاعی قارچ‌های *F. solani* و *M. phaseolina* را به ترتیب ۴۸/۳۶ درصد و ۶۴/۷ درصد مهار کرد. همچنین، *Saccharomyces cerevisiae* فعالیت ضدقارچی قابل توجهی از خود نشان داد؛ به‌ویژه در رقت پنجم، که میزان مهار رشد به ۷۴/۱ درصد برای *F. solani* و ۷۶/۱ درصد برای *M. phaseolina* رسید. این نتایج نشان داد که هر دو عامل زیستی در شرایط آزمایشگاهی به‌طور مؤثری رشد قارچ‌های بیماری‌زا را مهار می‌کنند. آزمون‌های بیماری‌زایی نیز تأیید کرد که آلودگی با *F. solani* و *M. phaseolina* موجب کاهش معنی‌دار درصد جوانه‌زنی بذر و افزایش مرگ‌ومیر گیاهچه‌ها می‌شود. با این حال، کاربرد عوامل کنترل زیستی شدت بیماری را به‌طور چشمگیری کاهش داد. در میان تمامی تیمارها، کاربرد همزمان *P. fluorescens* همراه با *M. phaseolina* بهترین نتیجه را به همراه داشت و در مقایسه با تیمارهای آلوده به عامل بیماری‌زا، موجب افزایش معنی‌دار درصد جوانه‌زنی و کاهش مرگ‌ومیر گیاهچه‌ها شد. نتایج آزمایش‌های گلخانه‌ای نیز نشان داد که *Saccharomyces cerevisiae* و *Pseudomonas fluorescens*، چه به‌صورت منفرد و چه به‌صورت ترکیبی، موجب افزایش معنی‌دار درصد جوانه‌زنی، میزان کلروفیل، رشد رویشی و سطح برگ شدند. افزون بر این، کاربرد توأم این دو عامل زیستی اثر هم‌افزایی قابل توجهی در مهار فعالیت عوامل بیماری‌زا، کاهش شیوع و شدت بیماری و بهبود رشد کلی گیاه از خود نشان داد.

نتیجه‌گیری: به‌طور کلی، نتایج این پژوهش نشان می‌دهد که *Saccharomyces cerevisiae* و *Pseudomonas fluorescens* عوامل کنترل زیستی امیدبخش و سازگار با محیط زیست هستند که می‌توان از آن‌ها به‌طور مؤثر در برنامه‌های مدیریت تلفیقی بیماری برای کنترل پوسیدگی ریشه بامیه ناشی از *Fusarium solani* و *Macrophomina phaseolina* استفاده کرد. این یافته‌ها از توسعه راهکارهای پایدار مدیریت بیماری حمایت می‌کنند؛ راهکارهایی که ضمن کاهش وابستگی به قارچ‌کش‌های شیمیایی، سلامت و بهره‌وری گیاه را نیز بهبود می‌بخشند.

کلمات کلیدی: عوامل کنترل زیستی، *Fusarium solani*، *Macrophomina phaseolina*، *Pseudomonas fluorescens*، *Saccharomyces cerevisiae*

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