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Research Article

Evaluation of Biogenically Synthesized Magnesium Nanoparticles Using Pumpkin Seed and *Agaricus bisporus* Extracts for the Management of White Rot Disease of Eggplant Caused by *Sclerotinia sclerotiorum*

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ABSTRACT

White rot caused by *Sclerotinia sclerotiorum* is an important disease that can substantially reduce crop growth and yield. The present study evaluated the antifungal and disease-suppressive potential of biosynthesized magnesium oxide nanoparticles (MgO-NPs) and *Agaricus bisporus* extracts against *S. sclerotiorum* and their effects on growth and yield of two cultivars (Jawhara and Barcelona). The most virulent fungal isolate obtained from infected plants was identified molecularly by PCR amplification of the ITS region, which produced an approximately 600-bp amplicon consistent with *S. sclerotiorum*. *In vitro* assays showed that MgO-NPs had the strongest antifungal activity, completely inhibiting mycelial growth at 0.70 mL L^{-1} and achieving 96% inhibition at 0.50 mL L^{-1}. The 5% alcoholic mushroom extract inhibited mycelial growth by 60%. SEM characterization revealed predominantly spherical MgO-NPs with particle sizes of approximately 20–50 nm and a mean diameter of about 35 nm. Under greenhouse conditions, the infected control exhibited the highest disease incidence (65.83%) and severity (0.638), whereas the 'alcoholic extract + MgO-NP' combined treatment completely suppressed disease incidence and reduced disease severity to 0.052. Treatment applications also substantially improved chlorophyll content, plant height, dry weight, and fruit production. The highest chlorophyll content (46.77 SPAD) was recorded with the' aqueous extract + MgO-NP' treatment, while MgO-NPs produced the highest dry weight (20.97 g) and mean fruit weight (2525.5 g $plant^{-1}$). Overall, biosynthesized MgO-NPs, particularly when combined with *A. bisporus* extracts, showed considerable potential for suppressing *S. sclerotiorum* and improving crop performance.

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Introduction

Eggplant (*Solanum melongena* L.), as a key vegetable crop, is widely grown in many parts of the world, including Iraq, and plays a significant role in the human diet and food security (FAO, 2023). Its production is, however, limited by a number of soil dwelling pathogens, including *Sclerotinia sclerotiorum*, the cause of white mold disease. The pathogen infects different plant tissues and may cause wilting, stem and root rots and finally mortality of plants, when the environmental conditions are conducive (Boland and Hall, 2021; Derbyshire and Denton-Giles, 2016). The ability of *S. sclerotiorum* to produce persistent sclerotia is an important mechanism for its survival in soil for longer periods when environmental conditions are

unfavorable. As a result, the disease outbursts significantly reduce plant growth and physiological functions, leading to significant yield losses and even total crop failure in extreme situations (Boland and Hall, 2021; Derbyshire and Denton-Giles, 2016).

Soil-borne fungal pathogens have been conventionally controlled by synthetic chemical fungicides. Even though they are effective, but their intensive and frequent applications can cause environmental pollution, result in the development of pathogen populations resistant to fungicides and have negative impacts on non-target species (Sharma et al., 2019, 2022). These constraints have necessitated the interest in conducting research to find out innocuous, ecofriendly and sustainable crop protection solutions. In this regard, the use of green nanotechnology as a possible approach to develop ecofriendly substitutes to traditional disease-management methods has become an interesting strategy (Khan et al., 2024). The utilization of magnesium based nanoparticles is of particular interest as magnesium is a vital plant nutrient and has a central role in chlorophyll molecule, and thus plays an important role in the fundamental photosynthesis process (Guo et al., 2022; Chrysargyris et al., 2023). Thus, biologically synthesized magnesium nanoparticles, could offer twofold benefits which is enhancement of plant growth and suppression of pathogens. They have been reported to possess antifungal properties which is linked to the production of reactive oxygen species (ROS), damage to fungal cell structures, and better plant tissue interactions (Khan et al., 2024; Abdelrhim et al., 2025).

Concurrently with developments in nanotechnology, mushroom-based products have gained substantial attention as part of sustainable plant disease-management systems. Edible mushrooms are a source of diverse biologically active compounds that could trigger plant defense reactions and help to increase tolerance to biotic and abiotic stresses (El Enshasy and Hatti-Kaul, 2022). *Agaricus bisporus* is a widely cultivated edible mushroom that contains high quantities of polysaccharides, especially β-glucans, phenolic compounds, vitamins, amino acids, proteins, and essential minerals (Feng et al., 2020; Sultan 2020). These compounds might be involved in various mechanisms that can promote plant growth and suppress diseases.

Mushroom-derived bioactive compounds can directly inhibit pathogenic microorganisms by affecting cell membrane integrity and cellular functions, while also stimulating plant defense responses through the accumulation of defense-related metabolites and activation of defense-associated enzymes (Wang et al., 2020). In addition, the antioxidant properties of mushroom extracts may help regulate oxidative stress and support plant growth and productivity. Collectively, these characteristics indicate that *A. bisporus* extracts could serve as promising natural products for enhancing plant health and suppressing phytopathogens (Ramos et al., 2019; Usman et al., 2021) Despite increasing interest in biologically synthesized nanoparticles and mushroom-derived products for sustainable crop protection, their combined application against *S. sclerotiorum* in eggplant has received little attention. To the best of our knowledge, no previous study has evaluated the combined efficacy of biologically synthesized magnesium nanoparticles derived from pumpkin seeds and aqueous or alcoholic extracts of *A. bisporus* against white mold disease of eggplant. Combining these two biological methods could yield complementary mechanisms of action and decrease synthetic fungicide use. Hence, the present study was conducted to assess the effectiveness of biologically synthesized magnesium nanoparticles from pumpkin seeds against *S. sclerotiorum* under laboratory and field situation. Moreover, the impact of these treatments was evaluated on vegetative growth and yield-related parameters of two eggplant cultivars 'Jawhara' and 'Barcelona'. The objective of the study was also to evaluate the efficacy and sustainability of these ecofriendly remedies for reducing white mold disease severity without compromising productivity of eggplant.

Materials and Methods

Laboratory experiments

Isolation and identification of pathogenic fungus

Eggplant plants with characteristic white mold disease symptoms were sampled and infected root and stem tissues were collected for identification and characterization of the pathogen. The samples were first washed thoroughly with water and surface sterilized with 3% sodium hypochlorite (NaOCl) for 2 min. The surface sterilized samples were cleaned with sterilized distilled water and transferred to autoclaved Potato Dextrose Agar (PDA) plates under sterilized conditions. The plates were kept at 25 ± 2°C for 5 days for fungal growth. Following incubation, the resultant fungal cultures were again grown on PDA to get purified colonies of *S. sclerotiorum* by hyphal tip method. The

purified isolates were subsequently transferred to fresh PDA plates and incubated under the same conditions to maintain actively growing, pure cultures of *S. sclerotiorum*.

Genomic DNA extraction

Genomic DNA was extracted from seven-day-old fungal cultures using the Chelex® 100 resin-based extraction method. Briefly, fungal mycelia were harvested from actively growing cultures and transferred aseptically into sterile microcentrifuge tubes containing 100 µL of Tris-EDTA (TE) buffer and 200 µL of Chelex® 100 resin suspension. The samples were heated in a boiling water bath for 10 min to facilitate cell lysis and DNA release. Following incubation, the samples were centrifuged at 16,000 rpm to pellet the Chelex® 100 resin and cellular debris. The resulting supernatant with genomic DNA was then carefully removed and transferred to new sterilized microcentrifuge tubes and directly subjected to further molecular processing.

DNA amplification by PCR

For molecular identification, universal primers ITS1 (5′-TCCGTAGGTGAACCTGCGG-3′) and ITS4 (5′-TCCTCCGCTTATTGATATGC-3′) were used to amplify the internal transcribed spacer (ITS) region of the ribosomal DNA of fungal isolates (White et al., 1990). Maxime PCR PreMix (i-Taq; Korea) was used for PCR amplification with a final reaction volume of 25 µL. The PCR reaction was performed under the following conditions: initial denaturation at 94°C for 3 min, followed by 35 cycles of denaturation at 94°C for 30 sec, annealing at 52°C for 45 sec and extension at 72°C for 45 sec. A final extension was carried out at 72°C for 7 min. The amplified products were subjected to electrophoresis in 1.5% agarose gel and observed to confirm the presence of the expected approximately 600 bp amplicon.

Preparation of aqueous *A. bisporus* extract

Thermal extraction method was used to prepare aqueous extracts of *A. bisporus*. A 5 g sample of fungal biomass was weighed and pulverized. The powdered material was dissolved in 50 mL distilled water and stirred continuously for 15 min at 60°C to extract the water soluble bioactive compounds. This mixture was then allowed to cool down at room temperature and first passed through cheese cloth and then filter paper to remove any particle matter. The filtrate obtained was instantly used for the experiments and biosynthesis of magnesium oxide nanoparticles (MgO-NPs) (Hassan et al., 2021).

Preparation of alcoholic *A. bisporus* extract

The dried *A. bisporus* powder was thoroughly homogenized by mixing with 70% ethanol at the proper ratio of powder to solvent. To improve extraction of bioactive compounds, the suspension was sonicated in an ultrasonic bath. After sonication, the solid residues were separated by filtration and the filtrate was concentrated through evaporation in a water bath for a period of 24 h. The concentrated extract was then dried at low temperature and kept in sterile airtight containers for future use (Umana et al., 2020).

Green synthesis of MgO-NPs

Using pumpkin seed extract as a biological stabilizing and capping agent, MgO-NPs were synthesized. In short, magnesium nitrate [$Mg(NO_3)_2$] was dissolved in 50 mL of distilled water while continuous stirring. Pumpkin seed extract was then added to the magnesium nitrate @ 50 mL and stirred continuously. Sodium hydroxide (NaOH; 2 g) was then added slowly to the reaction mixture with continuous stirring till the precipitation was observed.

The reaction mixture was kept in a water bath at 50–55°C for 24 h to allow the formation of nanoparticles. The resultant suspension was centrifuged for 15 min at 10,000 rpm and the precipitated material was retrieved. The precipitate was rinsed with distilled water three times and then with ethanol once to eliminate remaining reactants and other contaminants. After purification, the precipitate was dried in a hot-air oven for 4-8 h at 60°C. The resultant MgO-NPs were then moved to sterile, airtight containers, and stored until further use (Umaralikhan, 2018).

Characterization of MgO-NPs

The morphology and particle characteristics of the biosynthesized MgO-NPs were examined using scanning electron microscopy (SEM). Micrographs were obtained at a magnification of 20,000× using an accelerating voltage of 10.0 kV, a working distance of 5.5 mm, and SE2 (secondary electron) imaging mode. The SEM micrographs were analyzed using ImageJ software to estimate particle size and characterize the morphological features of the synthesized nanoparticles (Jeevanandam et al., 2017).

In vitro* evaluation of treatments against *S. sclerotiorum

The antifungal activity of the tested treatments against *S. sclerotiorum*, the causal agent of white mold, was evaluated using the poisoned food technique. PDA was prepared and sterilized by autoclaving. After cooling the medium to approximately 45–50°C, the respective treatments were incorporated into the medium, thoroughly mixed, and poured into sterile Petri dishes.

A 5-mm-diameter mycelial disc was excised from the actively growing margin of a 7-day-old culture of *S. sclerotiorum* and placed at the center of each PDA plate. Untreated PDA plates inoculated with the pathogen served as controls. The plates were incubated at 25 ± 2°C until the fungal colony in the control plates reached the edge of the Petri dish. Radial colony growth was then measured, and the inhibitory effect of each treatment was determined based on the reduction in fungal growth relative to the untreated control (Mueller et al., 2002).

Field experiment

A field experiment was conducted using a randomized complete block design (RCBD) to evaluate the efficacy of the treatments against *S. sclerotiorum* on two eggplant cultivars, Jawhara and Barcelona. The experimental area was divided into three blocks, with each block containing eight treatment plots. Each treatment was replicated three times, with each replicate considered an experimental unit containing eight plants.

The treatments were as follows:

T1 = Inoculated control (*S. sclerotiorum* only)

T2 = Healthy control (non-inoculated and untreated)

T3 = Alcoholic extract of *A. bisporus*

T4 = Aqueous extract of *A. bisporus*

T5 = Biosynthesized MgO-NPs

T6 = Combination of alcoholic and aqueous extracts of *A. bisporus*

T7 = Combination of alcoholic *A. bisporus* extract and MgO-NPs

T8 = Combination of aqueous *A. bisporus* extract and MgO-NPs

Inoculation with the pathogen

Eggplant plants were inoculated with *S. sclerotiorum* 90 days after transplanting. A 5-mm-diameter mycelial plug was excised from the actively growing margin of a PDA culture and placed on the stem approximately 10–15 cm above the soil surface. The inoculation site was covered with sterile cotton moistened with sterile distilled water and sealed with Parafilm to maintain adequate moisture and facilitate infection. Following inoculation, plants were maintained under high-humidity conditions to promote disease development (Gupta et al., 2020).

Treatment application

The treatments were first applied 7 days after inoculation with *S. sclerotiorum*. A second application was made 14 days after the first application. The treatments were applied as sprays directly to the infected portions of the plants and consisted of the alcoholic and aqueous extracts of *A. bisporus* and the biosynthesized MgO-NPs, either individually or in the respective combinations as described above.

Disease assessment

Disease incidence

Disease incidence was determined for each treatment by calculating the percentage of plants showing visible symptoms of *S. sclerotiorum* infection:

$$\text{Disease incidence (\%)} = \frac{\text{umber of infected plants}}{\text{Total number of plants assessed}} \times 100$$

Disease Severity

Disease severity was assessed using a visual rating scale based on the extent of disease development and tissue damage on stems and leaves. Disease severity was calculated using the formula of McKinney (1923):

$$\text{Disease severity (\%)} = \frac{\Sigma\ (\text{number of plants in each disease category} \times \text{Corresponding disease rating})}{\text{Total number of plants assessed} \times \text{Maximum disease rating}} \times 100$$

The disease severity scale consisted of seven categories as given in Table 1.

Chlorophyll content

Chlorophyll content of leaves was estimated by SPAD-502 chlorophyll meter. The SPAD value of each experimental unit was measured from three readings taken from representative leaves of each experimental unit and the average of the three readings was expressed as the SPAD value.

Average fruit weight

The fruits were picked at harvest from each experimental unit and weighed. Mean fruit weight was calculated and expressed in grams per fruit (g fruit^{-1}).

Total fruit yield

Fruits were harvested from each plant from each experimental unit over the harvest period and total fruit yield was estimated by weighing all the fruit with the help a balance. Values obtained were expressed as g plant^{-1}.

Statistical analysis

Analysis of variance (ANOVA) was used to analyze the experimental data, using SPSS statistical software. The effect of treatments, cultivars and treatment × cultivar was assessed. If significant differences were found, the treatment means were compared with one another at the 5% probability level ($P \leq 0.05$) by the least significant difference (LSD) test.

Table 1. Disease rating scale used for assessing disease severity.

Sr. No.	Disease Rating	Disease Severity
1	0	No infection
2	1	Infection ranging from 1–10%
3	2	Infection ranging from 11–25%
4	3	Infection ranging from 26–50%
5	4	Infection ranging from 51–75%
6	5	Infection ranging from 76–100%
7	6	Plant death

Results

Isolation and selection of the pathogen

Plant samples infected with white rot were taken from various sites such as Tikrit, Al-Alam, Baiji and Baghdad. Pathogen was isolated and cultured from diseased tissues on PDA culture media. The pathogenicity of all the fungal isolates was then tested on cabbage seeds and the most virulent isolate was chosen for further testing.

Molecular identification of the pathogen by PCR

The internal transcribed spacer (ITS) region, amplified by PCR with the primers ITS1 and ITS4, resulted in the formation of a distinct DNA band about 600 bp (Figure 1). The amplification profile from the fungal isolate was in agreement with the size of the ITS region and confirmed molecularly the fungal isolate as *Sclerotinia sclerotiorum*, the causal agent of white rot disease.

The amplicon size observed was similar to that reported previously, which indicated that the ITS region was a reliable molecular marker for the identification of *S. sclerotiorum* (Bolton et al., 2006).

In vitro inhibitory activity of biosynthesized MgO-NPs and *A. bisporus* extracts against *S. sclerotiorum*

The *in vitro* inhibitory activities of various concentrations of biosynthesized MgO-NPs and *A. bisporus* extracts against *S. sclerotiorum* are shown in Figures 2. The biosynthesized MgO-NPs showed the highest fungicidal activity. The mycelial growth was inhibited by 100% and 96% when exposed to the concentrations of 0.70 and 0.50 mL L^{-1} of MgO-NPs, respectively, resulting in colony diameters of 0.00 and 0.36 cm, respectively. The 5% alcoholic extract of mushroom showed the highest inhibitory activity with 60% mycelial growth inhibition. The untreated control only exhibited an inhibition 5.55%.

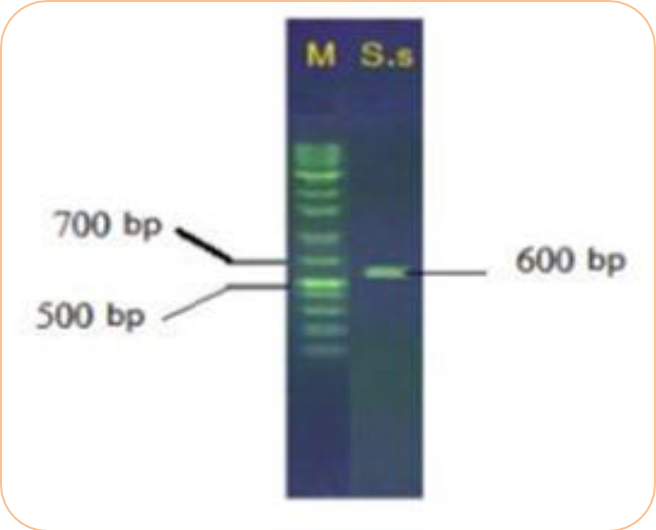

Figure 1. PCR amplification of *S. sclerotiorum* showing an approximately 600-bp DNA fragment. The PCR products were separated by electrophoresis on a 2% agarose gel in 1× TBE buffer at 5 V cm^{-1} for 1 h. N, 100-bp DNA ladder; M, molecular marker; S.s., *S. sclerotiorum*.

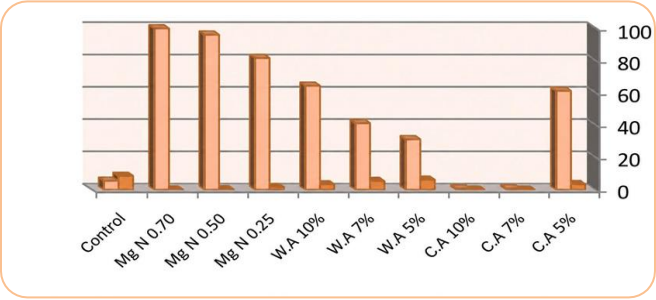

Figure 2. Inhibitory activity of different concentrations of biologically synthesized MgO-NPs and *A. bisporus* extracts against the growth of *S. sclerotiorum*. Mg N = Magnesium oxide nanoparticles (MgO-NPs), W.A = Aqueous extract of *A. bisporus*, and C.A = Alcoholic extract of *A. bisporus*.

Characterization of biosynthesized MgO-NPs

Scanning electron microscopy (SEM) was used to study the morphology and size of the biosynthesized nanoparticles. The SEM images at 20,000× magnification are shown in Figures 3. The synthesized particles were mostly in the nanoscale range with diameters of about 20–50 nm according to the 200 nm scale bar. The mean particle size was determined to be ~35 nm by image analysis with ImageJ. The particles were mainly spherical with some irregularities along the edges and with regard to overall dimension. These morphological

features confirmed the formation of nanoscale MgO particles through the biosynthesis process.

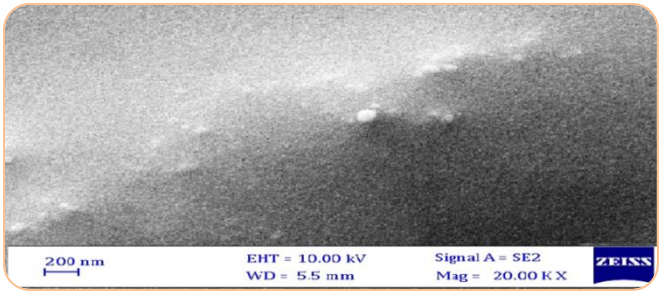

Figure 3. SEM image of biologically synthesized MgO-NPs at a magnification of 20,000×.

Effect of treatments on white rot disease incidence

Significant differences were observed in incidences of white rot disease among the treatments (Table 2). Disease incidence was the highest (65.83%) in the infected control in both the tested cultivars. In contrast, the healthy control, alcoholic extract, and MgO-NPs treatments recorded disease incidences of < 0.001%. The aqueous extract treatment resulted in a disease incidence of 15.83%, whereas the combined application of the aqueous extract and MgO-NPs resulted in 22.50% disease incidence. The combination of 'alcoholic extract and MgO-NPs' completely suppressed disease incidence with a recorded value of <0.001%.

Table 2. Effect of alcoholic and aqueous extracts of *A. bisporus*, biogenic MgO-NPs, and their combinations on the incidence of *S. sclerotiorum* of two eggplant cultivars.

Treatments	% Disease incidence		
	Jawhara cultivar	Barcelona cultivar	Mean
Infected control	68.33	63.33	65.83
Healthy control	<0.001	< 0.001	< 0.001
Alcoholic extract	<0.001	< 0.001	< 0.001
Aqueous extract	13.33	18.33	15.83
Biogenic MgO-NPs	< 0.001	< 0.001	< 0.001
Alcoholic extract+ Aqueous extract	20.00	25.00	22.50
Alcoholic extract + Biogenic MgO-NPs	< 0.001	5.00	2.50
Aqueous extract + Biogenic MgO-NPs	25.00	10.00	17.50
Cultivar Mean	15.83	15.21	15.52
L.S.D (0.05)	Treatments = 3.741	Cultivars = 1.870	Interactions = 5.290

Effect of treatments on white rot disease severity

Disease severity differed among treatments (Table 3). The infected control exhibited the highest disease severity, with a mean value of 0.638. In contrast, disease severity was reduced to <0.001 in the healthy control, alcoholic extract, and MgO-NPs treatments. The aqueous extract treatment resulted in a disease severity of 0.197, whereas the combined application of the alcoholic and aqueous extracts resulted in a severity value of 0.208. The combined 'alcoholic extract + MgO-NPs' treatment reduced disease severity to 0.052, while the 'aqueous extract + MgO-NPs' treatment resulted in a severity value of 0.130.

With respect to cultivar response, the cultivar Jawhara showed a mean disease severity of 0.155, while Barcelona cultivar showed a mean disease severity of 0.151, which indicated a relatively similar reaction of the two cultivars against *S. sclerotiorum* infection.

Effect of treatments on chlorophyll content

Table 4 shows the effect of treatments on chlorophyll content measured in terms of SPAD values. Application of treatments had a significant effect on the improvement of chlorophyll content in *S. sclerotiorum* challenged plants. The combined application of 'aqueous extract + biosynthesized MgO-NPs' was the most effective treatment which yielded the highest amount of chlorophyll content (46.77 SPAD), followed by the alcoholic extract treatment (45.30 SPAD). The chlorophyll content was the lowest in the infected control (20.17 SPAD) compared to other treatments. As far as the cultivar performance is concerned, Jawhara measured an average of 43.87 SPAD while Barcelona measured 39.16 SPAD.

Table 3. Effect of alcoholic and aqueous extracts of *A. bisporus*, biogenic MgO-NPs, and their combinations on disease severity of two eggplant cultivars.

Treatments	% Disease severity		
	Jawhara cultivar	Barcelona cultivar	Mean
Infected control	0.693	0.583	0.638
Healthy control	<0.001	<0.001	<0.001
Alcoholic extract	<0.001	<0.001	<0.001
Aqueous extract	0.180	0.213	0.197
Biogenic MgO-NPs	<0.001	<0.001	<0.001
Alcoholic extract+ Aqueous extract	0.187	0.230	0.208
Alcoholic extract + Biogenic MgO-NPs	0.043	0.060	0.052
Aqueous extract + Biogenic MgO-NPs	0.140	0.120	0.130
Cultivar Mean	0.155	0.151	0.153
L.S.D (0.05)	Treatments = 0.0928	Cultivars = 0.0464	Interactions = 0.1312

Table 4. Effect of alcoholic and aqueous extracts of *A. bisporus*, biogenic MgO-NPs, and their combinations on chlorophyll content (SPAD) of two eggplant cultivars challenged with *S. sclerotiorum*.

Treatments	Chlorophyll content (SPAD)		
	Jawhara cultivar	Barcelona cultivar	Mean
Infected control	20.17	20.17	20.17
Healthy control	66.60	62.00	64.30
Alcoholic extract	42.07	48.57	45.32
Aqueous extract	31.67	22.83	27.25
Biogenic MgO-NPs	50.27	40.17	45.22
Alcoholic extract+ Aqueous extract	42.50	33.13	37.82
Alcoholic extract + Biogenic MgO-NPs	44.87	45.73	45.30
Aqueous extract + Biogenic MgO-NPs	52.83	40.70	46.77
Cultivar Mean	43.87	39.16	41.52
L.S.D (0.05)	Treatments = 5.020	Cultivars = 2.510	Interactions = 7.099

Effect of treatments on plant height

The treatments significantly differed regarding plant height (Table 5). The 'alcoholic extract + biosynthesized MgO-NPs' conjoint treatment resulted in the maximum mean plant height of 46.45 cm followed by the treatment with MgO-NPs (44.95 cm). The height of infected control plants was the minimum with a mean of 29.25 cm. Regarding cultivars, Barcelona produced a significantly higher mean plant height of 43.17 cm compared to Jawhara (38.12 cm) cultivars.

Effect of treatments on plant dry weight

Significant differences in plant dry weight were observed among the treatments (Table 6). The MgO-NP-based treatment recorded a mean dry weight of 20.97 g, which was higher than that recorded under the other treatments. The combined 'alcoholic + aqueous extract' treatment resulted in a mean dry weight of 12.99 g, whereas the infected control recorded the lowest value (7.95 g). The healthy control had a mean dry weight of 21.25 g.

Regarding cultivar performance, Jawhara recorded a higher mean dry weight (16.34 g) than Barcelona (15.11 g). The treatment × cultivar interaction further showed that the MgO-NP-based treatment applied to Jawhara produced the highest dry weight (24.03 g).

Table 5. Effect of alcoholic and aqueous extracts of *A. bisporus*, biogenic MgO-NPs, and their combinations on plant height of two eggplant cultivars challenged with *S. sclerotiorum*.

Treatments	Plant height		
	Jawhara cultivar	Barcelona cultivar	Mean
Infected control	29.83	28.67	29.25
Healthy control	44.53	50.17	47.35
Alcoholic extract	34.70	49.10	41.9
Aqueous extract	32.57	35.43	34.0
Biogenic MgO-NPs	39.00	50.90	44.95
Alcoholic extract+ Aqueous extract	42.30	42.37	42.33
Alcoholic extract + Biogenic MgO-NPs	46.60	46.30	46.45
Aqueous extract + Biogenic MgO-NPs	35.47	42.40	38.93
Cultivar Mean	38.12	43.17	40.65
L.S.D (0.05)	Treatments = 2.230	1.115 = Cultivars	Interaction = 3.154

Table 6. Effect of alcoholic and aqueous extracts of *A. bisporus*, biogenic MgO-NPs, and their combinations on dry weight of two eggplant cultivars challenged with *S. sclerotiorum*.

Treatments	Dry weight		
	Jawhara cultivar	Barcelona cultivar	Mean
Infected control	7.87	8.03	7.95
Healthy control	22.60	19.90	21.25
Alcoholic extract	18.97	18.43	18.70
Aqueous extract	11.70	8.33	10.02
Biogenic MgO-NPs	24.03	17.90	20.97
Alcoholic extract+ Aqueous extract	11.00	14.97	12.99
Alcoholic extract + Biogenic MgO-NPs	22.33	18.83	20.58
Aqueous extract + Biogenic MgO-NPs	12.20	14.48	13.34
Cultivar Mean	16.34	15.11	15.72
L.S.D (0.05)	Cultivars = 0.789	Treatments = 1.579	Interaction = 2.233

Effect of treatments on mean fruit weight

Significant differences among treatments were also observed for mean fruit weight (Table 7). The MgO-NPs treatment resulted in the highest mean fruit weight (2525.5 g plant^{-1}), closely followed by the alcoholic extract treatment (2525.4 g plant^{-1}). In contrast, the infected control produced the lowest mean fruit weight (668.7 g plant^{-1}).

A significant difference was also observed between the cultivars. Jawhara produced a higher mean fruit weight (2611.0 g plant^{-1}) than Barcelona (2172.0 g plant^{-1}).

Discussion

The beneficial effects of biosynthesized MgO-NPs on vegetative growth observed in the present study may be associated with their nanoscale properties, which can influence nutrient availability, uptake, and utilization by plants. The small size of particles and high specific surface area of nanoparticles might allow them to interact with plant tissues and help to enhance the supply of Mg to plant cells. Magnesium is a vital macronutrient which is essential for plant growth and development and is involved in numerous physiological and biochemical processes. Apart from its nutritional role, the antimicrobial activity of nanoparticles containing magnesium has also been reported, indicating that magnesium-based nanoparticles can not only help plants to uptake nutrients but can also suppress the pathogens as well (Ali et al., 2024). These synergistic effects can lead to increased plant height, biomass production and general vegetative growth (Boone et al., 2023).

Table 7. Effect of alcoholic and aqueous extracts of *A. bisporus*, biogenic MgO-NPs, and their combinations on average fruit weight of two eggplant cultivars challenged with *S. sclerotiorum*.

Treatments	Average fruit weight		
	Jawhara cultivar	Barcelona cultivar	Mean
Infected control	852.3	485.0	668.7
Healthy control	2912.9	3085.2	2999.1
Alcoholic extract	2678.6	2372.2	2525.4
Aqueous extract	1692.7	1407.9	1550.3
Biogenic MgO-NPs	2819.2	2231.7	2525.5
Alcoholic extract+ Aqueous extract	2144.3	2049.5	2096.9
Alcoholic extract + Biogenic MgO-NPs	2265.8	2439.9	2352.8
Aqueous extract + Biogenic MgO-NPs	2010.0	2016.9	2013.4
Cultivar Mean	2172.0	2011.0	2091.5
L.S.D (0.05)	Cultivars = 39.17	Treatments = 78.35	Interaction = 110.80

The central atom of the chlorophyll molecule is magnesium and hence it is essential for the biosynthesis of chlorophyll and photosynthesis. There are also a number of enzymes that depend on sufficient Mg availability for their activities, which are involved in carbon assimilation, energy metabolism and other basic cellular functions (Zhang et al., 2024). Enhanced availability and maintenance of photosynthesis under pathogen pressure following MgO-NPs application in the present study, therefore, might be viewed as improvement of the chlorophyll content. Magnesium nutrition has also been linked to improved antioxidant defense mechanisms, such as activities of enzymes like catalases, superoxide dismutase, and peroxidases, which can help prevent the accumulation of excessive reactive oxygen species (ROS), thus reducing oxidative stress (Irshad et al., 2024). Therefore, under disease pressure, maintaining chloroplast and membrane integrity can help to sustain photosynthesis and plant growth.

Sufficient Mg supply might also help regulate carbohydrate metabolism and partitioning of assimilates between source and sink tissues. Magnesium also plays a role in phloem loading, carbohydrate transport and is possibly involved in osmotic regulation and cellular expansion (Wasule et al., 2024). Consequently, improved photosynthetic carbon assimilation and assimilate translocation could increase the availability of carbohydrates for biomass production and reproductive development. The greater dry weight and fruit production recorded in MgO-NP-treated plants in the present study may therefore reflect a combination of improved nutritional status, photosynthetic performance, and reduced pathogen-induced damage. Similar relationships between improved Mg nutrition, physiological performance, and biomass accumulation have been reported previously (Masood et al., 2024).

The application of *A. bisporus* extracts also improved plant performance and reduced disease development in the present study. The biological activity of *A. bisporus* extracts might be associated with their diverse array of bioactive constituents, including phenolic compounds, flavonoids, polysaccharides such as β-glucans, organic acids, amino acids, vitamins, and mineral elements (Al-Daraji, 2022). These compounds may exert both direct and indirect effects on plant health. Phenolic compounds and other secondary metabolites can possess antimicrobial and antioxidant properties, whereas polysaccharides and other biologically active compounds may stimulate plant defense responses. Collectively, these activities could reduce pathogen development and alleviate disease-associated oxidative stress, thereby maintaining cellular integrity and supporting plant growth.

The improved growth observed following application of *A. bisporus* extracts might also be related to their potential biostimulatory effects. Bioactive constituents in mushroom extracts can influence nutrient acquisition, enzymatic activity, and plant physiological processes, potentially promoting cell division, cell expansion, and biomass accumulation (Zhang et al., 2024). In plants challenged by *S. sclerotiorum*, such effects could be particularly important because reduced pathogen development would limit the diversion of plant resources towards defense and tissue repair.

Consequently, a greater proportion of photosynthetically derived assimilates may remain available for vegetative growth and reproductive development (Derbyshire and Denton-Giles, 2016).

The combined application of MgO-NPs and *A. bisporus* extracts produced particularly promising effects in several of the evaluated parameters. The enhanced performance of the combined treatments may result from complementary modes of action. MgO-NPs may exert direct antifungal effects, potentially through interactions with fungal cell membranes, disruption of cellular homeostasis, and induction of oxidative stress, whereas mushroom-derived bioactive compounds may contribute to pathogen inhibition while simultaneously stimulating plant defense and physiological processes. The combination of these activities could provide both direct suppression of *S. sclerotiorum* and indirect enhancement of host tolerance to infection. Such complementary effects may explain the improved plant growth and disease suppression observed with the combined treatments compared with some individual applications.

The *in vitro* results provide further support for the antifungal potential of the biosynthesized MgO-NPs. The highest concentration tested completely inhibited mycelial growth of *S. sclerotiorum*, whereas the mushroom extracts produced comparatively lower levels of inhibition. This difference indicates that MgO-NPs may have a stronger direct inhibitory effect on fungal growth under the tested laboratory conditions. However, the improved performance of some combined treatments under plant-based conditions suggests that direct antifungal activity alone may not fully explain disease suppression. Interactions among nanoparticle-mediated antifungal activity, the bioactive constituents of the mushroom extracts, and plant defense responses may collectively determine treatment efficacy.

The reduction in disease incidence and severity following MgO-NPs and *A. bisporus* treatments was accompanied by improved chlorophyll content, plant height, dry biomass, and fruit production. This relationship suggests that effective suppression of *S. sclerotiorum* reduced the negative effects of infection on plant physiological processes and allowed greater allocation of resources towards growth and reproduction. The markedly lower fruit production observed in the infected control further demonstrates the substantial effect of white rot on crop productivity. Importantly, the MgO-NPs treatment increased mean fruit production to 2525.5 g plant^{-1} compared with 668.7 g plant^{-1} in the infected control, indicating considerable potential for reducing the yield losses associated with *S. sclerotiorum* infection.

Although the present findings demonstrate promising biological activity, the proposed mechanisms should be interpreted cautiously because the underlying physiological and molecular responses were not directly measured. In particular, changes in antioxidant enzyme activity, reactive oxygen species, nutrient uptake, membrane integrity, fungal biomass, and plant defense-related gene expression were not quantified. Therefore, these mechanisms should be considered plausible explanations supported by previous literature rather than direct evidence generated by the present study. Further investigations incorporating physiological, biochemical, and molecular analyses are warranted to clarify the mechanisms responsible for the observed effects.

Conclusion

The present study demonstrated the potential of biosynthesized MgO-NPs and *A. bisporus* extracts for suppressing white rot caused by *S. sclerotiorum* and improving the growth and productivity of eggplant. MgO-NPs exhibited strong antifungal activity under *in vitro* conditions, while their application to infected plants was associated with reduced disease development and improved chlorophyll content, plant height, dry biomass, and fruit production. The combination of MgO-NPs with *A. bisporus* extracts also showed promising disease-management potential, indicating that integrating nanoparticle-based and biologically derived treatments may provide complementary benefits.

Among the evaluated treatments, MgO-NPs produced the highest mean fruit production (2525.5 g plant^{-1}), compared with 668.7 g plant^{-1} in the infected control. These findings suggest that biosynthesized MgO-NPs and mushroom-derived extracts could serve as promising components of more sustainable approaches for managing *S. sclerotiorum* in eggplant. However, their practical application should be evaluated further before being considered alternatives to conventional fungicides. In particular, treatment efficacy, optimal application rates, phytotoxicity, persistence, environmental effects,

and consistency under diverse production conditions require systematic assessment.

A limitation of the present study is that the experiments were conducted at a single location and during one growing season. In addition, comprehensive physicochemical characterization of the biosynthesized MgO-NPs using techniques such as transmission electron microscopy (TEM), X-ray diffraction (XRD), Fourier-transform infrared spectroscopy (FTIR), dynamic light scattering (DLS), zeta-potential analysis, and energy-dispersive X-ray spectroscopy (EDX) was not performed because these facilities were unavailable. Future research should therefore focus on detailed physicochemical characterization of the nanoparticles, determination of their stability and elemental composition, assessment of phytotoxicity and environmental safety, and evaluation of their effects on non-target organisms. Multi-location and multi-season field trials are also necessary to validate treatment efficacy, determine the consistency of disease suppression and yield responses, and establish the practical applicability of these treatments under diverse agricultural conditions.

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Author Contributions

The research plan was developed by MSJ, while the experimental and statistical design was developed and implemented by FMD. MSJ provided assistance with laboratory equipment and contributed to the molecular diagnosis of the fungus. FMD prepared the first draft of the manuscript, while MSJ critically reviewed, proofread, and approved the final version for publication.

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Conflict of Interest

The authors declare no conflict of interest.

Sustainable Development Goals Targeted

SDG 2: Zero Hunger
SDG 12: Responsible Consumption and Production
SDG 15: Life on Land

Policy Addressed

The findings support policies promoting sustainable crop protection, integrated disease management, reduced dependence on synthetic fungicides, and environmentally responsible nanotechnology. They also highlight the need for regulatory frameworks addressing nanoparticle safety and environmental fate, while supporting resilient vegetable production and enhanced food security through effective, sustainable disease management.

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