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### Biocontrol of *Rhizoctonia solani* Induced Root Rot in Sugar Beet via Organic Inputs and *Chaetomium globosum*: Greenhouse and Field Evaluation

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#### ABSTRACT

*Rhizoctonia solani* poses a significant threat to sugar beet production in Egypt, causing considerable stand losses. A study, involving 27 isolates from various governorates, revealed variable virulence, with post-emergence-damping-off ranging from 10-35%. The most aggressive isolate, RHI1, causing 35% damping-off, was selected for further evaluation. The research focused on an integrated pest management strategy combining compost with the biocontrol fungus *Chaetomium globosum*. Greenhouse trials demonstrated a remarkable reduction in disease incidence. Post-emergence-damping-off plummeted to 5.53% in treated plants, a significant decrease from 45.37% in untreated controls. Disease severity also fell drastically, from 65.33 to 8.37%. Results were confirmed in field trials under natural infection, where disease incidence was minimized to 10.2% in treated plots compared to 62.7% in controls. Beyond disease suppression, integrated treatment significantly enhanced vegetative growth. Treated plants in greenhouse trials exhibited superior growth parameters, including more leaves (18.67 vs. 12.33), increased height (31.83cm vs. 18.20cm), and higher fresh and dry weights. Similar improvements in yield and bulb weight were observed in field conditions. The study further elucidated the mechanisms behind these improvements. Biochemical assays revealed a strong induction of defense-related enzymes in treated plants, with elevated peroxidase and polyphenol oxidase activity. Additionally, soil microbiological analysis indicated a healthier soil ecosystem in treated plots, with increased populations of beneficial fungi, bacteria, and actinomycetes. This study supports the integration of compost and *C. globosum* as an effective, eco-friendly alternative to chemical fungicides for sustainable sugar beet cultivation, offering significant disease suppression (over 70%), enhance plant growth, and improved soil health.

**Keywords:** *Rhizoctonia solani*, (*Beta vulgaris*), Biocontrol, Organic amendments, Enzyme activity (POD & PPO)

#### INTRODUCTION

Sugar beet (*Beta vulgaris* L.) is a commercially significant crop cultivated primarily in temperate regions due to its high sucrose content. It ranks as the second most important global source of sugar after sugarcane, contributing approximately 20% to worldwide sugar production (Subrahmanyeswari and Gantait, 2022). Major producers include Russia, the United States, Germany, France, and Turkey (RAGT Seeds, 2024). In Egypt, sugar beet is the second most vital sugar-producing crop following sugarcane (Fouad and Shaker, 2025). Its value extends beyond sugar production, serving as a feedstock for bioethanol, animal feed, and raw material for various biotechnological industries, thereby enhancing its role in food security and renewable energy (FAO, 2023).

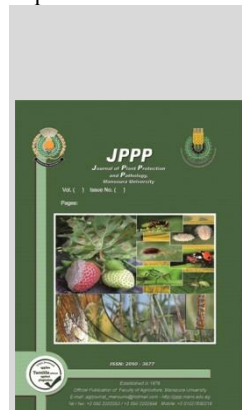
Despite its economic and agricultural importance, sugar beet productivity is severely affected by soil-borne diseases, particularly root and crown rots caused by *Rhizoctonia solani* and *Sclerotium rolfsii*. These pathogens can lead to yield losses ranging from 30% to complete crop failure under severe conditions (El Mansoub *et al.*, 2020 and Misra *et al.*, 2023). Such diseases are especially problematic in heavy, poorly drained soils under warm and moist conditions, resulting in plant death, reduced root quality, and diminished sugar content (Haque and Parvin, 2021).

Traditional disease control methods—such as crop rotation, early planting, and chemical fungicides—have shown inconsistent results. Moreover, excessive fungicide use raises concerns about environmental safety, human health, and the

emergence of resistant pathogen strains (Lamichhane *et al.*, 2017, Farhaoui *et al.*, 2022 and Alqahtani, 2025).

Consequently, sustainable alternatives are gaining attention. These include organic soil amendments (e.g., compost and crop residues) and biological control agents such as *Trichoderma* spp., *Chaetomium globosum*, yeasts, and *Streptomyces*, which enhance soil microbial activity, stimulate plant defense enzymes like peroxidase (POD) and polyphenol oxidase (PPO), and suppress pathogenic fungi (El-Tarabily, 2004, Misra *et al.*, 2023 and Rehman *et al.*, 2024).

However, most studies have examined these methods in isolation, with limited data on their combined application under field conditions. This underscores the need for integrated disease management (IDM) strategies that combine organic amendments, biocontrol agents, and reduced chemical inputs. Such approaches not only improve disease suppression but also enhance soil fertility, reduce reliance on agrochemicals, and support sustainable agriculture. Therefore, the present study aims to evaluate the combined effects of compost, various straw types, and beneficial biocontrol agents on soil microbial communities, defense enzyme activities, vegetative growth, and disease suppression in sugar beet under both greenhouse and field conditions. This integrated approach is expected to offer effective and sustainable solutions for managing root and crown rot while improving crop productivity and reducing dependence on chemical fungicides.



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## MATERIALS AND METHODS

### Study Site and Experimental Design

This study was conducted at the Faculty of Agriculture, Ain Shams University, Cairo, Egypt. Laboratory and greenhouse experiments were designed to evaluate the management of sugar beet (*Beta vulgaris* L.) root rot caused by *R. solani*. To validate the efficacy of the treatments under natural conditions, field trials were carried out in a naturally infested site located in El-Gosq village, Sharqia Governorate, Egypt.

### Plant Material

Seeds of sugar beet (*Beta vulgaris* L., cv. *Clavius*) were obtained from the Agricultural Research Center, Giza, Egypt.

### Isolation, Identification, and Inoculum Preparation of *Chaetomium globosum*

*Chaetomium globosum* was isolated from the rhizosphere of sugar beet plants. The isolate was purified and identified based on morphological characteristics, including the structure of mature perithecia, terminal hairs, asci, and ascospores, following the descriptions provided by Walther and Gindrat (2011). Inoculum was prepared by inoculating sterilized 500 mL bottles containing a mixture of straw and sorghum grains with 5 mm discs from 7-day-old fungal cultures. The bottles were incubated at 27 °C for three weeks, following the protocol of Sharma *et al.* (2025).

### Preparation of Plant Residues

Post-harvest residues of rice (*Oryza sativa*), maize (*Zea mays*), and quinoa (*Chenopodium quinoa*) were collected from local farms. The residues were manually cleaned to remove soil and debris, rinsed with tap water, and shade-dried at ambient temperatures (25–30 °C) for 7–10 days to stabilize moisture content. Direct sunlight was avoided to preserve nutrient composition and structural integrity (Yu *et al.*, 2024).

Dried residues were cut into 3–5 cm segments using a mechanical cutter and sieved through a 10 mm mesh to ensure uniform particle size. Prior to soil incorporation, residues were treated as follows:

1. Moisture content was adjusted to approximately 65% of water-holding capacity using clean water.
2. Urea was added at a rate of 0.5% (w/w) of dry residue weight to optimize the carbon-to-nitrogen (C: N) ratio and enhance microbial decomposition (Yu *et al.*, 2024).
3. For treatments involving *C. globosum*, 15-day-old colonized inoculum (incubated at 25 ± 2 °C) was added at 1% (w/w) and thoroughly mixed with the moistened residues.

The treated materials were incubated under shaded ambient conditions for seven days to promote partial microbial colonization prior to application.

**Note:** The applied urea concentration is considered safe for both sugar beet (*B. vulgaris* var. *saccharifera*) and *C. globosum*, if incorporation occurs at least 14 days prior to planting in greenhouse trials and 21 days prior to planting in field trials.

### Compost Amendment

The compost used in greenhouse and field experiments was supplied by Prof. Dr. Fawzy Abo El-Abass (Faculty of Agriculture, Ain Shams University). It was produced under aerobic conditions from a mixture of plant residues and animal manure. The compost was fully decomposed, dark brown in color, and exhibited an earthy odor. Prior to application, it was sieved through a 10 mm mesh to ensure homogeneity (Schumann *et al.*, 2023). The physicochemical properties of compost are presented in Table 1.

**Table 1. Chemical and physical analysis of the compost amendment applied in the study**

Analysis	Organic amendments
Weight per cubic meter (kg)	560
Moisture %	12
pH (1:10)	7.67
EC (1:10) dS/m	9.97
Total N %	1.86
N-NH <sub>4</sub> (ppm)	361
N-NO <sub>3</sub> (ppm)	18
Organic material %	44.25
Organic carbon %	25.66
Ash %	55.75
Total P %	0.49
Total K %	1.46
C/N	1: 13.79
weed seeds	-

“The chemical and physical analyses of compost were carried out at the Soils, Water and Environment Research Institute (SWERI), Ministry of Agriculture and Land Reclamation, Egypt. Abbreviations: N = nitrogen, P = phosphorus, K = potassium, C = carbon.”

### Application of the Fungicide Folicure

In this experiment, Folicure® 25% EC (tebuconazole 25%) was employed as a chemical control reference to evaluate the efficacy of the tested soil amendments and biological agents. The fungicide was procured from the Ministry of Agriculture, Egypt.

### Laboratory Experiment: Fungal Frequency in Sugar Beet Root Rot

#### 1. Isolation and Identification of Fungi

Sugar beet seedlings showing damping-off symptoms were collected from fields in Sharqia Governorate. Diseased root tissues were surface sterilized using 5% sodium hypochlorite for 2 minutes, then air-dried and plated on potato dextrose agar (PDA) medium. Plates were incubated at 25 °C for five days.

Emerging fungal colonies were purified and identified based on morphological characteristics, following the methods described by Farhaoui *et al.* (2023). Pure cultures were maintained on PDA slants, incubated at 25 °C for 7 days, and stored at 5 °C for future use.

#### 2. Frequency Calculation of Fungal Isolates

The frequency of each fungal species was calculated using the formula described by Mohamed *et al.* (2018):

$$\text{Frequency of Pathogen \%} = \frac{\text{Number of samples in which the fungus occur}}{\text{Total number of examined sample}} \times 100$$

### Greenhouse Experiments:

#### 1. Pathogenicity Test:

Each fungal isolate was cultured separately in 500 mL glass bottles containing 200 g of sterilized barley grains, which support optimal mycelial growth. The inoculated bottles were incubated at 28–30 °C for 14 days (Felsőciová *et al.*, 2021). Sterilized sandy-clay soil was infested with the prepared inoculum at a rate of 2% (w/w) one week before sowing. Control pots contained sterilized soil only. Ten seeds of the sugar beet cultivar *Clavius* (poly-germ) were sown per pot, and seedling emergence was recorded and the incidence of pre-emergence and post-emergence damping-off was quantified using the formula:

$$\text{Damping-off (\%)} = \frac{\text{Number of infected plants}}{\text{Total number of plants}} \times 100$$

This approach aligns with prior evaluations conducted at 15 days (pre-emergence) and 45 days (post-emergence) after planting, as reported in sugar beet damping-off studies (Abbas, *et al.* 2022).

Following disease quantification, the pathogen causing the highest severity was identified as *R. solani*. This conclusion was based on both its morphological characteristics in culture and the manifestation of typical root-rot symptoms in sugar beet seedlings during pathogenicity assays, reflecting its well-documented virulence and prevalence as a principal causal agent in sugar beet seedling diseases (Abbas, *et al.* 2022 and Wibberg *et al.* 2016).

## 2. Greenhouse Experimental Design

A pot experiment was carried out under greenhouse conditions to evaluate the effect of different organic and biological amendments on sugar beet root-rot caused by *R. solani*. The treatments were arranged in a randomized complete block design (RCBD) with three replicates. Each treatment was applied to sterilized pots filled with artificially infested soil, and sugar beet seeds were sown under controlled greenhouse conditions.

Sterilized pot, 30 cm in diameter, was filled with 5 kg sterilized soil was artificially inoculated with *R. solani* at 2% (w/w) using barley grain inoculum. Treatments were applied as follows:

- **Single amendment:** 2% w/w (100 g per 5 kg soil).
- **Amendment + C.g:** 2% straw + 1% C.g inoculum (w/w).
- **Compost only:** 2% w/w (100 g per 5 kg soil).
- **Compost + C.g:** 2% compost + 1% C.g (w/w).

Amendments were thoroughly incorporated into infested soil 14 days before planting. Soil moisture was maintained at ~60% water-holding capacity throughout the pre-plant incubation period.

Ten seeds of sugar beet were seeded in each pot, 30 cm in diameter and 5 pots were used for each treatment as replicates as well as the control. Un-amended infested pots were used as control.

Both disease parameters (post-emergence damping-off, disease incidence and disease severity) and vegetative growth traits Plant (height, leaf number, fresh and dry weights) Root (length, diameter, fresh and dry weights) were recorded to evaluate treatment efficacy.

### Disease Assessment

Root rot incidence and severity were assessed at harvest using the 0–7 rating scale proposed by Campbell *et al.*, (2014), where:

- 0 = no visible lesions,
- 1 = arrested lesions at the point of inoculation,
- 2 = <5% shallow, dry rot canker,
- 3 = 5–24% deep, dry rot canker,
- 4 = 25–49% extensive rot,
- 5 = 50–89% rot penetrating into the root interior,
- 6 = 90 to < 100% rot with most foliage dead,
- 7 = 100% plant mortality.

$$\text{Disease severity (\%)} = \frac{\sum (\text{rating no.}) \times (\text{no. roots in rating category}) \times (100)}{(\text{Total no. roots}) \times (\text{highest rating value})}$$

## Field Experiments:

### 1. Field Experimental Design.

The experiments were carried out at El-Gosaq village, Sharqia governorate. The experiment was conducted in a naturally *R. solani*-infested field. Land preparation involved ploughing and leveling, followed by uniform broadcasting of the prepared amendments and incorporation into the top 15 cm of soil:

- **Single amendment:** 5 ton/ha (2.1 ton/Fadden).

- **Amendment + C.g:** 5 ton/ha (2.1 ton/Fadden) straw + 2.5 ton/ha (1.1 ton/Fadden) C.g inoculum.
- **Compost only:** 5 ton/ha (2.1 ton/Fadden).
- **Compost + C.g:** 5 ton/ha (2.1 ton/Fadden) compost + 2.5 ton/ha (1.1 ton/Fadden) C.g inoculum.

Incorporation took place 21 days before planting, with immediate irrigation to stimulate decomposition. Soil moisture was kept close to field capacity until planting.

All treatments as well as non-treated control were replicated three times in a randomized complete block design and each consisted of plots each 1 m in width, 7 m length and 50 cm apart. Planting was 50 cm apart using double ridges on each row.

Both disease parameters (post-emergence damping-off, disease incidence and disease severity) and vegetative growth traits Foliar (length (cm), weights(gm)) Root (length (cm), weights(gm)) were recorded to evaluate treatment efficacy.

### 2. Impact of Treatments on Soil Microbial Populations

The populations of rhizospheric microorganisms, including total bacteria, fungi, and actinomycetes, were quantified two weeks after the application of treatments. The standard serial dilution plate method, originally described by Sreevidya *et al.* (2016), was employed to assess microbial abundance. Specific selective media were used for each microbial group: soil extract agar for total bacterial counts, potato dextrose agar supplemented with rose Bengal for fungal counts, and oatmeal agar for actinomycetes.

Soil suspensions were serially diluted and plated onto the respective media, followed by incubation at 27 °C under different durations depending on the microbial group: 2 days for bacteria, 3 days for actinomycetes, and 4 days for fungi. After incubation, colony-forming units (CFUs) were enumerated using a digital colony counter, and results were expressed as CFU per gram of oven-dry soil.

### 4. Peroxidase and Polyphenol Oxidase Activities

Fresh plant tissues were homogenized in ice-cold 100 mM potassium phosphate buffer (pH 7.0) supplemented with 0.1 mM EDTA and 1% polyvinylpyrrolidone (PVP, w/v) to prevent phenolic oxidation. The homogenates were centrifuged at 15,000 × g for 15 min at 4 °C, and the resulting supernatants were used as crude enzyme extracts.

Peroxidase (POD; EC 1.11.1.7) activity was determined following the method of Hammerschmidt *et al.* (1982) and Abo-Zaid (2020) using guaiacol as substrate, and absorbance changes were recorded at 470 nm. One unit of POD activity was defined as an increase of 0.01 in absorbance min<sup>-1</sup>.

Polyphenol oxidase (PPO; EC 1.14.18.1) activity was assayed according to Benjamin and Montgomery (1973) and Abo-Zaid (2020) by monitoring absorbance at 420 nm, where one unit was defined as a change of 0.001 absorbance min<sup>-1</sup>.

Protein concentration in the extracts was quantified using the Gabr *et al.* (2020) method with bovine serum albumin as standard. Enzyme activities were expressed as units per mg protein, and all measurements were conducted using a UV-Vis spectrophotometer (UV 9100 B, LabTech, China).

### 5. Statistical Analysis

The obtained data were analyzed using Analysis of Variance (ANOVA) following the General Linear Model (GLM) procedure described in the *SAS User's Guide* (SAS Institute, 2012). Treatment means were compared using

Duncan's Multiple Range Test, and statistical significance was determined at the 5% probability level (Duncan, 1955).

## RESULTS AND DISCUSSIONS

### Results

#### 1. Laboratory Experiments:

##### Frequency of fungi associated with Sugar beet root rot plants:

Analysis of fungal isolates associated with sugar beet root rot symptoms (Fig. 1) revealed that *R. solani* was the most frequently detected species (27.27%), suggesting its potential dominance as the primary pathogen under the conditions surveyed. *F. solani* followed with a frequency of 18.2%, while *F. oxysporum* and *F. semitectum* were each identified in 13.63% of samples (Fig. 2). In contrast, *Pythium* spp., *Sclerotium rolfsii*, and *Macrophomina phaseolina* were less common, each accounting for 9.1% of isolates. These results indicate that *R. solani* may play a central role in disease development, with other fungi contributing to a lesser but still notable extent.

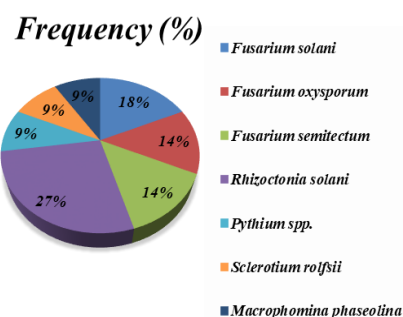


Fig. 1. Frequency of fungi associated with Sugar beet root rot plants.



Fig. 2. Samples of sugar beet infected with root rot disease.

#### 2. Greenhouse Experiments:

##### Pathogenicity Test:

Pathogenicity assays revealed considerable variation among the tested fungal isolates in their ability to cause disease under greenhouse conditions. *R. solani* isolates (Fig. 3) exhibited post-emergence damping-off incidences ranging from 10% to 35%, with isolate RH1 recording the highest percentage (35%), while all isolates showed no pre-emergence damping-off. *Pythium* spp. (Fig. 5) displayed a wide variation, as PYTH2 caused the highest post-emergence incidence among all tested isolates (60%), whereas PYTH1 resulted in 35%. For *S. rolfsii*, (Fig. 5) isolate SCL.R1 caused 35% post-emergence damping-off, while SCL.R2 showed the highest pre-emergence incidence (60%) but only 10% post-emergence.

Regarding *Fusarium* species (Fig. 4), *F. solani* isolates caused post-emergence damping-off ranging from 10% to 35%, with FS1 and FS3 reaching the upper limit (35%). *F. oxysporum* showed strong pathogenicity, particularly isolate FO1, which caused 55% post-emergence incidence, while FO3 showed the lowest among its group (15%). Similarly, *F. semitectum* isolates varied from 15% to 35%, with FSE3 showing relatively high incidence (30%).

*M. phaseolina* (Fig. 5) exhibited moderate virulence, with post-emergence incidence ranging from 10% to 25%. Notably, all fungal isolates failed to cause any pre-emergence damping-off in the autoclaved soil control, where both pre- and post-emergence incidences were zero.

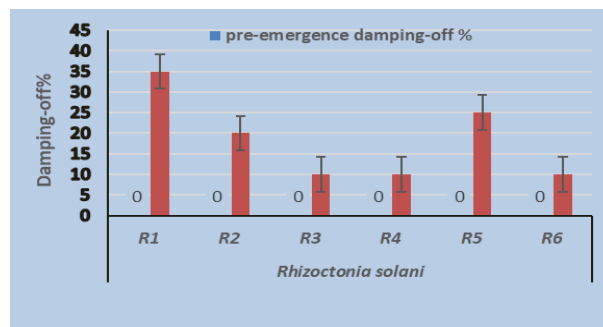


Fig. 3. Pathogenicity of *R. solani* Isolates on Sugar Beet under Greenhouse Conditions.

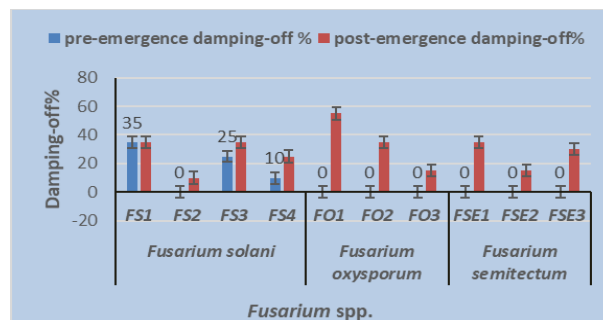


Fig. 4. Pathogenicity of *Fusarium* spp. Isolates on Sugar Beet under Greenhouse Conditions

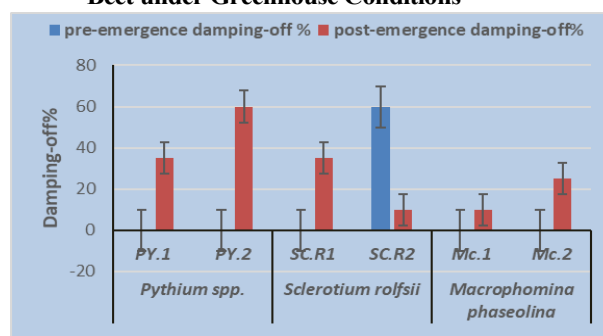


Fig.5. Pathogenicity of *Pythium* spp.; *Sc. rolfsii* and *M. phaseolina* Isolates on Sugar Beet under Greenhouse Conditions.

##### Greenhouse Experimental Design

The obtained results (Table 2) revealed that all tested treatments significantly reduced post-emergence damping-off, disease incidence, and disease severity of sugar beet compared to the infested control. The fungicide treatment (Folicure) recorded the lowest values of disease incidence (5.00%) and disease severity (17.33%), followed by the combined application of *C. globosum* with compost, which



showed 10.53% post-emergence damping-off, 12.50% disease incidence, and 18.17% disease severity. Similarly, *C. globosum* combined with quinoa straw ranked third in effectiveness, achieving 12.03% post-emergence damping-off and 19.27% disease severity.

Among organic amendments alone, compost exhibited the best performance (20.00%, 26.63%, and 26.83% for damping-off, incidence, and severity, respectively), while rice straw and maize straw showed moderate control efficiency, with disease severity values of 33.00% and 32.00%, respectively. Quinoa straw alone

resulted in slightly lower disease severity (31.07%) compared to rice and maize straw.

In contrast, the control treatment with infested soil recorded the highest values for post-emergence damping-off (35.00%), disease incidence (56.27%), and disease severity (65.67%), confirming the high disease pressure in the experiment. The autoclaved soil control showed no disease symptoms across all parameters, indicating that the observed disease was entirely due to the presence of soilborne pathogens.

**Table 2. Effect of Treatments on post-emergence damping-off and Disease Incidence and Severity of Sugar Beet under Greenhouse conditions.**

Treatment	post-emergence damping-off%	Disease Incidence %	Disease Severity
<i>Chaetomium globosum</i>	17.07 <sup>E</sup> ±0.23	20.00 <sup>F</sup> ±1.44	25.07 <sup>E</sup> ±0.15
+ Compost	10.53 <sup>F</sup> ±0.35	12.50 <sup>H</sup> ±1.44	18.17 <sup>L</sup> ±0.20
+ Rice straw	14.97 <sup>E</sup> ±0.15	20.00 <sup>F</sup> ±1.44	23.40 <sup>F</sup> ±0.12
+ maize straw	15.30 <sup>E</sup> ±0.46	23.63 <sup>E</sup> ±0.73	22.07 <sup>G</sup> ±0.34
+ Quinoa straw	12.03 <sup>F</sup> ±0.14	16.10 <sup>G</sup> ±1.81	19.27 <sup>H</sup> ±0.34
Compost	20.00 <sup>D</sup> ±0.23	26.63 <sup>CD</sup> ±0.44	26.83 <sup>D</sup> ±0.22
Rice straw	25.03 <sup>B</sup> ±0.15	32.50 <sup>CB</sup> ±1.44	33.00 <sup>B</sup> ±0.23
maize straw	24.00 <sup>CB</sup> ±0.06	33.67 <sup>B</sup> ±0.73	32.00 <sup>CB</sup> ±0.12
Quinoa straw	21.97 <sup>CD</sup> ±0.15	30.00 <sup>CD</sup> ±1.44	31.07 <sup>C</sup> ±0.30
Folicure	10.10 <sup>F</sup> ±0.21	5.00 <sup>L</sup> ±1.44	17.33 <sup>L</sup> ±0.43
Control infested soil	35.00 <sup>A</sup> ±2.89	56.27 <sup>A</sup> ±0.72	65.67 <sup>A</sup> ±0.95
Control autoclaved soil	0.00 <sup>G</sup> ±0.00	0.00 <sup>L</sup> ±0.00	0.00 <sup>L</sup> ±0.00

Data presented as the means of three replicates ± SD. Different letters refer to significant difference ( $P \leq 0.05$ ). L.S.D. at 0.05%

The obtained results (Table, 3) demonstrated that all tested treatments significantly enhanced root growth parameters of sugar beet compared to the infested untreated control. The combined application of *C. globosum* with compost recorded the highest values for root diameter (23.19 cm), root length (19.00 cm), root fresh weight (22.13 g), and root dry weight (1.83 g), surpassing all other treatments. Similarly, *C. globosum* combined with quinoa straw achieved superior performance, particularly in root diameter (22.84 cm) and fresh weight (20.80 g). In contrast, the infested untreated control exhibited the lowest growth, with root diameter, length, fresh weight, and dry weight reaching only 7.67 cm, 10.67 cm, 3.73 g, and 0.19 g, respectively. Autoclaved soil control maintained moderate values, while treatments with compost or organic amendments alone showed variable but generally lower improvements compared to their combinations with *C. globosum*. Application of the chemical fungicide Folicure resulted in slight increases over the infested control but remained markedly inferior to most bio-organic combinations (Fig. 6).



**Fig. 6. Effect of Different Treatments (T1: *C. globosum*, T2: C.g+Compost, T3: C.g+ Rice straw, T4: C.g+ maize straw, T5: C.g+ Quinoa straw, T6: Compost, T7: Rice straw, T8: maize straw, T9: Quinoa straw, CF: Folicure, CP: Control infested soil, CN: Control autoclaved soil) on Sugar Beet Disease Severity on Root and Foliar Growth under Greenhouse conditions.**

**Table 3. Effect of Different Treatments on Sugar Beet Root Growth Parameters under Greenhouse conditions.**

Treatment	Root Growth Parameters			
	Diameter(cm)	Length (cm)	Fresh weight (gm)	Dry weight (gm)
<i>C. globosum</i>	16.95 <sup>CB</sup> ±0.01	13.33 <sup>D</sup> ±0.022	12.27 <sup>E</sup> ±0.23	0.73 <sup>D</sup> ±0.03
+ Compost	23.19 <sup>A</sup> ±0.01	19 <sup>A</sup> ±0.29	22.13 <sup>A</sup> ±0.31	1.83 <sup>A</sup> ±0.03
+ Rice straw	22.42 <sup>A</sup> ±0.29	14.67 <sup>C</sup> ±0.27	13.37 <sup>D</sup> ±0.30	0.72 <sup>D</sup> ±0.01
+ maize straw	17.89 <sup>B</sup> ±0.01	15.33 <sup>CB</sup> ±0.60	16.17 <sup>C</sup> ±0.19	0.95 <sup>C</sup> ±0.003
+ Quinoa straw	22.84 <sup>A</sup> ±0.01	16.33 <sup>B</sup> ±0.44	20.80 <sup>B</sup> ±0.06	1.35 <sup>B</sup> ±0.10
Compost	16.04 <sup>CB</sup> ±3.20	13.33 <sup>D</sup> ±0.17	9.33 <sup>GF</sup> ±0.12	0.65 <sup>D</sup> ±0.01
Rice straw	15.42 <sup>CB</sup> ±0.01	12.17 <sup>EF</sup> ±0.33	8.60 <sup>G</sup> ±0.30	0.51 <sup>E</sup> ±0.01
maize straw	15.32 <sup>CB</sup> ±0.34	11.83 <sup>EF</sup> ±0.17	3.77 <sup>L</sup> ±0.29	0.47 <sup>E</sup> ±0.04
Quinoa straw	15.49 <sup>CB</sup> ±0.01	12.33 <sup>ED</sup> ±0.44	7.30 <sup>H</sup> ±0.61	0.21 <sup>GF</sup> ±0.02
Folicure	10.39 <sup>ED</sup> ±0.75	11.60 <sup>EF</sup> ±0.55	4.20 <sup>L</sup> ±0.06	0.24 <sup>GF</sup> ±0.01
Control infested soil	7.67 <sup>E</sup> ±0.01	10.67 <sup>G</sup> ±0.18	3.73 <sup>L</sup> ±0.39	0.19 <sup>G</sup> ±0.02
Control autoclaved soil	13.36 <sup>CD</sup> ±2.96	11.17 <sup>FG</sup> ±0.17	9.63 <sup>F</sup> ±0.09	0.31 <sup>F</sup> ±0.01

Data presented as the means of three replicates ± SD. Different letters refer to significant difference ( $P \leq 0.05$ ). L.S.D. at 0.05%

Data in Table (4) show that all applied treatments significantly improved the vegetative growth parameters of sugar beet plants compared to the untreated infested control. The combination of *C. globosum* with compost recorded the highest values for number of leaves (18.67), plant height (31.83 cm), fresh weight (25.30 g), and dry weight (1.83 g), followed closely by *C. globosum* combined with quinoa straw (18.33 leaves, 25.20 cm, 24.50 g, and 1.71 g, respectively). Treatments involving *C. globosum* with rice straw or maize straw also showed significant improvements over the control,

though to a lesser extent. In contrast, organic amendments applied alone, particularly maize and quinoa straw, resulted in moderate growth enhancement, whereas the chemical fungicide Folicure produced relatively lower growth metrics compared to bio-organic combinations. The lowest values for all growth parameters were observed in the infested untreated control (11.33 leaves, 18.23 cm, 7.80 g fresh weight, and 0.37 g dry weight), while the autoclaved soil control exhibited moderate performance, reflecting the absence of pathogen pressure.

**Table 4. Effect of Different Treatments on Sugar Beet Foliar Growth Parameters under Greenhouse conditions.**

Treatment	Foliar Growth Parameters			
	Number of leaves	Length (cm)	Fresh weight (gm)	Dry weight (gm)
<i>C. globosum</i>	16.0 <sup>B</sup> ±0.58	22.70 <sup>DC</sup> ±0.52	18.80 <sup>C</sup> ±0.25	1.23 <sup>D</sup> ±0.06
+ Compost	18.67 <sup>A</sup> ±0.33	31.83 <sup>A</sup> ±0.69	25.30 <sup>A</sup> ±0.68	1.83 <sup>A</sup> ±0.06
+ Rice straw	16.67 <sup>B</sup> ±0.33	23.37 <sup>C</sup> ±0.58	21.00 <sup>B</sup> ±0.25	1.64 <sup>B</sup> ±0.06
+ maize straw	16 <sup>B</sup> ±1.00	23.50 <sup>C</sup> ±1.00	20.33 <sup>B</sup> ±0.35	1.44 <sup>C</sup> ±0.09
+ Quinoa straw	18.33 <sup>A</sup> ±0.33	25.20 <sup>B</sup> ±0.25	24.50 <sup>A</sup> ±0.31	1.71 <sup>BA</sup> ±0.08
Compost	15.67 <sup>B</sup> ±0.33	21.33 <sup>DE</sup> ±0.60	13.77 <sup>D</sup> ±0.35	0.84 <sup>F</sup> ±0.03
Rice straw	13.33 <sup>C</sup> ±0.33	20.50 <sup>E</sup> ±0.29	13.87 <sup>D</sup> ±0.03	0.58 <sup>FG</sup> ±0.03
maize straw	13.33 <sup>C</sup> ±0.67	20.50 <sup>E</sup> ±0.29	10.67 <sup>E</sup> ±0.12	0.73 <sup>FE</sup> ±0.01
Quinoa straw	13.33 <sup>C</sup> ±0.88	20.00 <sup>F</sup> ±0.58	10.67 <sup>E</sup> ±0.17	0.83 <sup>E</sup> ±0.01
Folicure	11.67 <sup>DC</sup> ±0.33	20.50 <sup>E</sup> ±0.29	9.53 <sup>F</sup> ±0.12	0.41 <sup>HG</sup> ±0.08
Control infested soil	11.33 <sup>D</sup> ±0.33	18.23 <sup>F</sup> ±0.43	7.80 <sup>G</sup> ±0.21	0.37 <sup>H</sup> ±0.03
Control autoclaved soil	13.33 <sup>C</sup> ±0.33	21.27 <sup>DE</sup> ±0.43	13.10 <sup>D</sup> ±0.40	0.50 <sup>HG</sup> ±0.05

Data presented as the means of three replicates ± SD. Different letters refer to significant difference ( $P \leq 0.05$ ). L.S.D. at 0.05%

### 3. Field Experiments:

#### Field Experimental Design.

Under naturally infested field conditions, all tested treatments significantly reduced post-emergence damping-off, disease incidence, and disease severity of sugar beet compared to the untreated control (Table 5 and Fig. 7). The untreated control recorded the highest values for post-emergence damping-off (38.43%), disease incidence (84.17%), and disease severity (41.50%), which were statistically superior to all other treatments.

Among the evaluated treatments, Folicure application exhibited the greatest efficacy, resulting in the lowest post-emergence damping-off (3.80%), disease incidence (15.03%), and disease severity (10.90%), with highly significant differences compared to the control. This was followed by *C. globosum* combined with compost, which also provided notable disease suppression, recording 4.73% damping-off, 21.07% incidence, and 14.73% severity. Similarly, *C. globosum* alone and its combinations with rice straw or maize straw resulted in a marked decrease in all disease parameters, with damping-off values ranging from 5.23% to 5.70% and severity values between 15.37% and 21.40%.

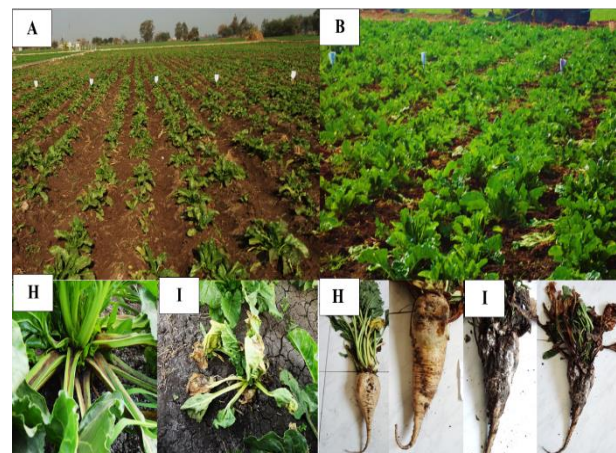
Organic amendments alone, such as compost, rice straw, maize straw, or quinoa straw, were generally less effective than their combinations with *C. globosum*. Compost alone reduced damping-off to 6.67% and severity to 20.27%, while rice straw and maize straw alone recorded higher damping-off percentages (10.43% and 18.30%, respectively) and correspondingly higher severity levels. Quinoa straw alone showed intermediate performance, with damping-off at 8.10% and severity at 29.40%.

Overall, chemical treatment with Folicure achieved the highest disease suppression, followed by integrated applications of *C. globosum* with organic amendments, whereas sole application of plant residues was less effective.

**Table 5. Effect of Treatments on post-emergence damping-off and Disease Incidence and Severity of Sugar Beet under Field conditions.**

Treatment	post-emergence damping-off%	Disease Incidence %	Disease Severity
<i>C. globosum</i>	5.53 <sup>C</sup> ±0.44	24.77 <sup>A</sup> ±0.15	15.53 <sup>C</sup> ±0.35
+ Compost	4.73 <sup>C</sup> ±0.20	21.07 <sup>B</sup> ±0.30	14.73 <sup>C</sup> ±0.27
+ Rice straw	5.23 <sup>C</sup> ±0.20	24.7 <sup>C</sup> ±0.20	15.37 <sup>C</sup> ±0.41
+ maize straw	5.7 <sup>C</sup> ±0.20	21.27 <sup>B</sup> ±0.52	21.4 <sup>BD</sup> ±0.26
+ Quinoa straw	7.267 <sup>BD</sup> ±0.28	21.07 <sup>B</sup> ±0.09	18.67 <sup>E</sup> ±0.27
Compost	6.67 <sup>EE</sup> ±0.23	26.1 <sup>C</sup> ±0.17	20.27 <sup>E</sup> ±0.37
Rice straw	10.43 <sup>C</sup> ±0.44	37.1 <sup>B</sup> ±0.21	22.23 <sup>D</sup> ±0.18
maize straw	18.3 <sup>B</sup> ±0.32	36.93 <sup>B</sup> ±0.29	24.3 <sup>C</sup> ±0.47
Quinoa straw	8.1 <sup>D</sup> ±0.36	37.17 <sup>B</sup> ±0.17	29.4 <sup>B</sup> ±0.37
Folicure	3.8 <sup>F</sup> ±0.17	15.03 <sup>E</sup> ±0.17	10.9 <sup>F</sup> ±0.15
Control untreated soil	38.43 <sup>A</sup> ±1.06	84.17 <sup>A</sup> ±2.62	41.5 <sup>A</sup> ±0.76

Data presented as the means of three replicates ± SD. Different letters refer to significant difference ( $P \leq 0.05$ ). L.S.D. at 0.05%



**Fig. 7. Effect of Treatments on Sugar Beet under Field conditions. A: general image of field experiment, B: close up image of some treatment, H: healthy plant, I: infected plant.**

Under naturally infested field conditions, significant differences were observed among treatments in terms of foliar length, foliar fresh weight, root length, and root fresh weight of sugar beet (Table 6). The untreated control exhibited the lowest growth performance, with foliar length of 40.00 cm, foliar fresh weight of 219.67 g, root length of 31.33 cm, and root fresh weight of 213.00 g.

The combined application of *C. globosum* with compost resulted in the highest enhancement of plant growth, recording the greatest foliar length (61.67 cm), foliar fresh weight (1235.00 g), root length (57.33 cm), and root fresh weight (1852.33 g), with highly significant differences compared to all other treatments. *C. globosum* combined with quinoa straw ranked second, particularly in root length (51.67 cm) and root fresh weight (1355.33 g), followed by combinations with rice straw or maize straw, which also showed notable improvement in growth parameters relative to the control.

The application of *C. globosum* alone enhanced growth compared to the control but was less effective than its combinations with organic amendments. Organic amendments applied alone generally produced lower values compared to integrated treatments, with compost outperforming rice straw, maize straw, and quinoa straw in most parameters.

The chemical treatment with Folicure did not substantially improve growth parameters, showing values close to the lower range of the tested treatments, and remained far below the integrated biocontrol-organic approaches.

Overall, integrating *C. globosum* with compost produced the most pronounced improvement in both foliar and root growth traits, followed by combinations with plant residues, while sole applications of organic materials or chemical treatment were less effective.

**Table 6. Effect of Different Treatments on Sugar Beet Growth Parameters under Field conditions.**

Treatment	Foliar Length (cm)	Foliar Fresh weight (gm)	Root Length (cm)	Root Fresh weight (gm)
<i>C. globosum</i>	49.67 <sup>C</sup> ±0.88	760.33 <sup>E</sup> ±0.88	40 <sup>D</sup> ±1.15	1337.67 <sup>C</sup> ±1.45
+ Compost	61.67 <sup>A</sup> ±0.88	1235 <sup>A</sup> ±1.15	57.33 <sup>A</sup> ±0.88	1852.33 <sup>A</sup> ±1.45
+ Rice straw	50.33 <sup>C</sup> ±0.88	1075.67 <sup>B</sup> ±1.20	42 <sup>D</sup> ±1.15	1165.33 <sup>E</sup> ±1.45
+ maize straw	50 <sup>C</sup> ±1.15	951.33 <sup>C</sup> ±0.8	48 <sup>C</sup> ±1.15	1275.33 <sup>D</sup> ±1.45
+ Quinoa straw	56 <sup>B</sup> ±1.15	832 <sup>D</sup> ±1.15	51.67 <sup>B</sup> ±1.20	1355.33 <sup>B</sup> ±1.45
Compost	49.67 <sup>C</sup> ±0.88	627.67 <sup>G</sup> ±1.45	40 <sup>D</sup> ±1.15	1012.33 <sup>F</sup> ±1.45
Rice straw	44.33 <sup>D</sup> ±0.88	547.33 <sup>H</sup> ±1.76	39.33 <sup>ED</sup> ±1.45	740 <sup>I</sup> ±0.58
maize straw	44.33 <sup>D</sup> ±0.88	542.33 <sup>I</sup> ±1.45	36.33 <sup>EF</sup> ±1.45	835.33 <sup>G</sup> ±1.45
Quinoa straw	46.67 <sup>D</sup> ±0.88	706.33 <sup>F</sup> ±0.88	39 <sup>ED</sup> ±1.15	753 <sup>H</sup> ±2.08
Folicure	46 <sup>D</sup> ±1.15	522.33 <sup>J</sup> ±1.45	35.33 <sup>F</sup> ±0.88	692.33 <sup>J</sup> ±1.45
Control untreated soil	40 <sup>E</sup> ±1.15	219.67 <sup>K</sup> ±0.88	31.33 <sup>G</sup> ±0.88	213 <sup>K</sup> ±1.15

Data presented as the means of three replicates ± SD. Different letters refer to significant difference ( $P \leq 0.05$ ). L.S.D. at 0.05%

#### Impact of Treatments on Soil Microbial Populations

Under naturally infested soil conditions, all tested treatments significantly influenced the total microbial counts compared to the untreated control (Table 7). The highest fungal counts were recorded in soils treated with *C. globosum* either alone or in combination with organic amendments, with values ranging from 40.30 to 42.50  $\times 10^6$  CFU g<sup>-1</sup> soil. The

combination of *C. globosum* with compost achieved the highest bacterial population (111.00  $\times 10^5$  CFU g<sup>-1</sup>), followed by its combination with quinoa straw (54.20  $\times 10^5$  CFU g<sup>-1</sup>) and rice straw (51.90  $\times 10^5$  CFU g<sup>-1</sup>).

**Table 7. Effect of Different Treatments on Total Microbial Counts in Soil (Fungi, Bacteria, and Actinomycetes) under naturally infested soil with *R. solani*.**

Treatments	Total count (CFU)		
	colony of fungi (106/g-1)	colony of Bacteria (105/g-1)	colony of actinomycetes (105/g-1)
<i>C. globosum</i>	40.30 <sup>A</sup> ± 0.30	41.63 <sup>E</sup> ± 0.65	10.60 <sup>F</sup> ± 0.65
+ Compost	42.50 <sup>A</sup> ± 0.50	111.00 <sup>A</sup> ± 1.00	15.57 <sup>A</sup> ± 0.85
+ Rice straw	41.50 <sup>A</sup> ± 0.50	51.90 <sup>C</sup> ± 0.56	14.40 <sup>B</sup> ± 0.20
+ maize straw	42.17 <sup>A</sup> ± 0.76	45.00 <sup>D</sup> ± 0.36	12.57 <sup>C</sup> ± 0.25
+ Quinoa straw	42.40 <sup>A</sup> ± 0.61	54.20 <sup>B</sup> ± 0.80	15.43 <sup>A</sup> ± 0.25
Compost	39.83 <sup>B</sup> ± 0.45	46.03 <sup>D</sup> ± 0.46	10.83 <sup>E</sup> ± 0.42
Rice straw	36.43 <sup>C</sup> ± 0.45	40.53 <sup>E</sup> ± 0.65	10.80 <sup>E</sup> ± 0.50
maize straw	32.70 <sup>D</sup> ± 0.30	38.33 <sup>F</sup> ± 0.80	11.77 <sup>D</sup> ± 0.25
Quinoa straw	36.27 <sup>C</sup> ± 0.76	38.07 <sup>F</sup> ± 0.95	11.60 <sup>D</sup> ± 0.30
Folicure	10.33 <sup>F</sup> ± 0.25	20.13 <sup>H</sup> ± 0.80	4.82 <sup>G</sup> ± 0.22
Control untreated soil	14.57 <sup>E</sup> ± 0.72	23.27 <sup>G</sup> ± 0.30	7.20 <sup>F</sup> ± 0.26

Data presented as the means of three replicates ± SD. Different letters refer to significant difference ( $P \leq 0.05$ ). L.S.D. at 0.05%

Actinomycetes populations were also markedly increased by integrated treatments, with *C. globosum* + compost (15.57  $\times 10^5$  CFU g<sup>-1</sup>) and *C. globosum* + quinoa straw (15.43  $\times 10^5$  CFU g<sup>-1</sup>) showing the highest counts. In contrast, organic amendments applied alone produced moderate increases in microbial populations, with compost recording 10.83  $\times 10^5$  CFU g<sup>-1</sup> actinomycetes and rice straw 10.80  $\times 10^5$  CFU g<sup>-1</sup>, while maize straw and quinoa straw alone yielded slightly higher actinomycete counts (11.77 and 11.60  $\times 10^5$  CFU g<sup>-1</sup>, respectively).

The chemical fungicide Folicure significantly reduced all microbial groups, with the lowest fungal (10.33  $\times 10^6$  CFU g<sup>-1</sup>), bacterial (20.13  $\times 10^5$  CFU g<sup>-1</sup>), and actinomycete (4.82  $\times 10^5$  CFU g<sup>-1</sup>) counts. The untreated control also exhibited low microbial populations, though higher than Folicure, indicating the suppressive effect of the fungicide on beneficial soil microbiota.

Overall, the integration of *C. globosum* with compost was the most effective treatment in enhancing total microbial populations in soil, followed closely by combinations with

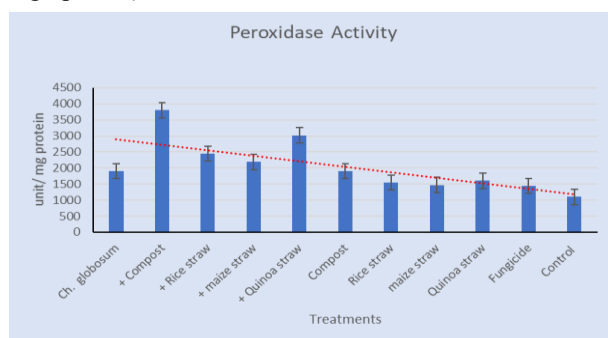


plant residues, while chemical treatment reduced microbial abundance.

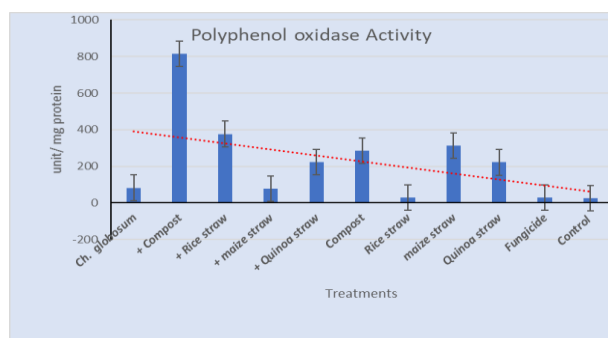
### Peroxidase and Polyphenol Oxidase Activities

Both peroxidase (POD) and polyphenol oxidase (PPO) activities showed significant enhancement under different organic and biological amendments compared with the untreated control.

For POD activity (Fig. 8), the highest value was recorded in soil amended with compost in combination with *C. globosum* (3804.40 U mg<sup>-1</sup> protein), followed by quinoa straw amendment (3018.23 U mg<sup>-1</sup> protein), and rice straw combined with *C. globosum* (2447.85 U mg<sup>-1</sup> protein). Moderate increases were observed with maize straw combined with *C. globosum* (2188.49 U mg<sup>-1</sup> protein) and compost alone (1902.28 U mg<sup>-1</sup> protein). The lowest activity was recorded in the untreated control (1105.15 U mg<sup>-1</sup> protein) and fungicide treatment (1442.66 U mg<sup>-1</sup> protein). For PPO activity (Fig. 9), the application of compost combined with *C. globosum* resulted in the maximum specific activity (815.14 U mg<sup>-1</sup> protein), followed by rice straw amendment (376.79 U mg<sup>-1</sup> protein) and maize straw alone (313.21 U mg<sup>-1</sup> protein). Intermediate levels were observed with quinoa straw alone (221.88 U mg<sup>-1</sup> protein) and compost alone (284.79 U mg<sup>-1</sup> protein). In contrast, the lowest PPO activity was recorded in the untreated control (25.94 U mg<sup>-1</sup> protein) and the fungicide treatment (28.62 U mg<sup>-1</sup> protein).



**Fig. 8.** Effect of organic, biological, and chemical treatments on peroxidase (POD) specific activities (unit mg<sup>-1</sup> protein) in sugar beet plants under Field conditions.



**Fig. 9.** Effect of organic, biological, and chemical treatments on polyphenol oxidase (PPO) specific activities (unit mg<sup>-1</sup> protein) in sugar beet plants under Field conditions.

Overall, the results indicate that organic amendments, particularly when integrated with the antagonistic fungus *C. globosum*, substantially enhanced both POD and PPO activities compared with chemical treatment or untreated

control. This suggests that such treatments may play an important role in activating host defense mechanisms against soil-borne pathogens.

### Discussion

The analysis of fungal isolates from sugar beet roots exhibiting root rot symptoms revealed that *R. solani* was the most prevalent pathogen, accounting for 27.27% of the isolates. This finding corroborates with previous studies identifying *R. solani* as a dominant causal agent of sugar beet root rot, particularly under conditions of elevated soil moisture and temperature that enhance its pathogenic activity.

Following *R. solani*, *Fusarium solani* was the second most frequently isolated species, representing 18.2% of the total. This result aligns with findings from Morocco, where *F. solani* was reported as the most aggressive *Fusarium* species affecting sugar beet, with disease incidences ranging from 37.5% to 100% (Chenaoui *et al.*, 2017).

*F. oxysporum* and *F. semitectum* were isolated at a frequency of 13.63%. *F. oxysporum* is known to cause Fusarium yellows, a disease characterized by leaf yellowing and wilting due to vascular tissue colonization. Although less frequently documented, *F. semitectum* has also been associated with sugar beet root rot (Burlakoti *et al.*, 2010; Chenaoui *et al.*, 2017).

Lower isolation frequencies were observed for *Pythium* spp., *Sclerotium rolfsii*, and *M. phaseolina*, each at 9.1%. *Pythium* spp. is recognized for causing pre- and post-emergence damping-off and root rot in various crops, including sugar beet. *S. rolfsii* is a soil-borne pathogen responsible for southern blight, which can affect sugar beet roots under favorable environmental conditions. *M. phaseolina* is notable for its broad host range and its capacity to induce root rot in sugar beet (Abbas *et al.*, 2022).

The predominance of *R. solani* in this study underscores its significant role in sugar beet root rot under the prevailing environmental conditions. However, the detection of multiple fungal species, even at lower frequencies, suggests a complex disease etiology potentially involving synergistic interactions among pathogens. Such interactions may exacerbate disease severity and complicate management strategies.

These findings emphasize the need for integrated disease management approaches that account for the diversity of fungal pathogens involved in sugar beet root rot. Recommended strategies include crop rotation, the use of resistant cultivars, and targeted fungicide applications tailored to the specific pathogen profile of the region (Misra *et al.*, 2023).

The pathogenicity assays revealed significant variability among the tested fungal isolates in their capacity to induce damping-off under controlled greenhouse conditions. *R. solani* isolates demonstrated post-emergence disease incidences ranging from 10% to 35%, with isolate RHI1 reaching the highest level (35%). These findings are consistent with prior reports identifying *R. solani* as a major soil-borne pathogen responsible for post-emergence damping-off in sugar beet, particularly under conditions of high humidity and moderate temperatures (Misra, *et al.* 2023 and Farhaoui, *et al.* 2022). The absence of pre-emergence damping-off among *R. solani* isolates aligns with its typical mode of infection, which predominantly affects young seedlings after emergence.

*Pythium* spp. displayed a wide range of virulence, with isolate PYTH2 inducing the highest post-emergence incidence



(60%) and PYTH1 causing 35%. *Pythium* species are well-documented as highly aggressive pathogens causing both pre- and post-emergence damping-off in sugar beet and other crops, often favored by saturated soils (Weiland, *et al.* 2015). Interestingly, the higher post-emergence incidence suggests that the isolates tested may have reduced pre-emergence activity under the greenhouse conditions employed, possibly due to environmental factors or inoculum distribution.

*Sclerotium rolfsii* isolates exhibited contrasting behavior: SCL.R1 caused 35% post-emergence damping-off, whereas SCL.R2 caused 60% pre-emergence but only 10% post-emergence incidence. *S. rolfsii* is known to produce sclerotia that survive in soil and infect seedlings both pre- and post-emergence, with disease expression strongly influenced by inoculum density and soil temperature (Chowdary, *et al.* 2024). The differential behavior among isolates underscores the genetic and virulence variability within *S. rolfsii* populations.

Among *Fusarium* species, *F. solani* isolates caused post-emergence incidences ranging from 10% to 35%, with FS1 and FS3 reaching the upper limit. *F. oxysporum* isolates displayed strong pathogenicity, particularly FO1, which induced 55% post-emergence incidence, whereas FO3 caused only 15%. Similarly, *F. semitectum* isolates varied from 15% to 35%, with FSE3 showing relatively high incidence (30%). These results are in agreement with studies reporting that *Fusarium* species are significant contributors to sugar beet damping-off, with pathogenicity often strain-specific and influenced by environmental conditions (Abdelghany, *et al.* 2024).

*M. phaseolina* exhibited moderate virulence, with post-emergence incidences ranging from 10% to 25%. This pathogen, known for causing charcoal rot in a wide range of crops, generally has lower aggressiveness on sugar beet seedlings compared to *R. solani* or *Pythium* spp., but it can exacerbate disease under stress conditions such as drought or high soil temperatures (Marquez, *et al.* 2021).

Overall, these results highlight the complex etiology of sugar beet damping-off, with multiple pathogens contributing to disease severity in varying degrees. The variability in virulence among isolates of the same species suggests that effective management strategies should consider pathogen diversity, environmental conditions, and cultivar susceptibility to reduce the impact of damping-off in sugar beet cultivation.

The greenhouse experiments demonstrated that all tested treatments significantly mitigated post-emergence damping-off, disease incidence, and disease severity of sugar beet compared to the untreated infested control. The chemical fungicide Folicure was the most effective treatment, reducing disease incidence to 5.00% and disease severity to 17.33%. This aligns with previous reports highlighting the high efficacy of systemic tebuconazole fungicides in suppressing soil-borne pathogens such as *R. solani* in sugar beet seedlings (Farhaoui, *et al.* 2022).

Among the biocontrol and organic amendments, the combined application of *C. globosum* with compost exhibited strong disease suppression, achieving 10.53% post-emergence damping-off, 12.50% disease incidence, and 18.17% disease severity. This is consistent with evidence that *C. globosum* can antagonize soilborne pathogens through mechanisms such as mycoparasitism, antibiotic production, and induction of systemic resistance, while compost enhances soil microbial diversity and suppresses pathogen proliferation (Sharma, *et al.* 2025). Similarly, the combination of *C. globosum* with quinoa straw resulted in substantial disease

reduction, highlighting the synergistic potential of integrating antagonistic fungi with organic amendments.

Organic amendments applied alone also contributed to disease suppression, albeit less effectively than the combined treatments. Compost alone reduced post-emergence damping-off, incidence, and severity to 20.00%, 26.63%, and 26.83%, respectively. Rice and maize straw achieved moderate control with disease severity around 32–33%, whereas quinoa straw alone was slightly more effective (31.07%). These observations support previous findings that organic residues can improve soil structure, promote beneficial microbial communities, and reduce pathogen activity, though their efficiency depends on the type of residue, decomposition rate, and nutrient content (Huang, *et al.* (2025) and Arcand, *et al.* 2016).

The untreated infested soil control exhibited the highest disease levels, with 35.00% post-emergence damping-off, 56.27% disease incidence, and 65.67% severity, confirming the high pathogenic pressure present in the experimental soil. Conversely, the autoclaved soil control showed no symptoms, verifying that disease expression was solely due to soilborne pathogens and eliminating the influence of abiotic factors.

Overall, these results indicate that integrating biocontrol agents like *C. globosum* with organic amendments can substantially reduce sugar beet damping-off, and such strategies may provide environmentally sustainable alternatives to chemical fungicides. The combination of organic and biological approaches appears particularly promising for managing complex soilborne pathogen populations while promoting soil health and crop resilience (Bonanomi, *et al.* 2020 and Walther, D. and Gindrat, D. 2011).

The enhancement of root growth parameters observed in sugar beet under *R. solani* infection indicates the efficacy of bio-organic and chemical treatments in mitigating pathogen-induced stress. The combined application of *C. globosum* with compost significantly promoted root diameter, length, fresh weight, and dry weight, surpassing both the individual organic amendments and chemical fungicide treatments. This synergistic effect can be attributed to the dual action of the antagonistic fungus and the nutrient-rich organic substrate. *C. globosum* is known for producing antifungal metabolites, cell wall-degrading enzymes, and plant growth-promoting compounds, which collectively suppress pathogen activity and enhance host vigor (Yu, *et al.* 2025 and El-Hadidy, 2018).

Organic amendments, such as compost and quinoa straw, provide additional benefits by improving soil physicochemical properties, increasing microbial diversity, and enhancing nutrient availability, which contribute to improved root development (Chaudhury, *et al.* 2023).

The superior performance of combined treatments over individual amendments aligns with previous studies reporting that bio-organic formulations enhance biocontrol efficacy and plant growth more than single applications of either biocontrol agents or organic matter alone (Tyagi, *et al.* 2024).

The decomposition dynamics of the applied residues were particularly influential in shaping the microbial environment. Corn straw, characterized by a relatively lower lignin content, decomposed faster and released simple sugars and organic acids that provided a readily available carbon source for antagonistic microbes such as *Chaetomium* and *Trichoderma* (Wozniak, *et al.* 2021). In contrast, rice straw, with its higher lignin and silica content, decomposed more slowly but released phenolic compounds and soluble silica,

which not only improved soil structure but also enhanced plant defense mechanisms by reinforcing cell walls (Tahaa, *et al.* 2021).

Interestingly, quinoa straw, which contains higher levels of nitrogen and protein compared to cereal residues, decomposed rapidly and released nitrogenous compounds that stimulated the proliferation of beneficial microbial communities. This could explain the stronger induction of systemic resistance and suppression of *Rhizoctonia* observed in quinoa straw treatments, aligning with earlier evidence that high-protein residues accelerate microbial colonization and pathogen suppression (Chaudhury, *et al.* 2023). On the other hand, compost amendment provided a more stable and balanced improvement in soil health through the release of humic and fulvic acids and the adjustment of soil pH. These changes created favorable conditions for beneficial microbial populations, while humic substances themselves acted as biostimulants that enhanced plant defense enzyme activities (Ghorbani, *et al.* 2008 and Bonanomi, *et al.* 2018).

While chemical fungicide application (Folicure) did slightly increase root parameters compared to the infested untreated control, its effectiveness was lower than the bio-organic combinations. This observation reflects the limitation of chemical treatments in improving plant growth under biotic stress, as they primarily target pathogen suppression without contributing to soil health or microbial-mediated growth promotion (Wójtowicz, *et al.* 2022).

Overall, the results demonstrate that integrating antagonistic fungi with organic amendments not only effectively suppresses *R. solani* but also significantly enhances root development in sugar beet, highlighting the importance of bio-organic strategies in sustainable crop management.

The enhancement of vegetative growth parameters in sugar beet under *R. solani* infection highlights the efficacy of bio-organic treatments in mitigating pathogen-induced stress. The combination of *C. globosum* with compost achieved the highest improvements in number of leaves, plant height, and biomass accumulation, closely followed by *C. globosum* with quinoa straw. These findings suggest a synergistic interaction between the antagonistic fungus and organic amendments, which not only suppress pathogen activity but also improve nutrient availability and soil structure, thereby promoting plant growth (Yu, *et al.* 2025 and El-Hadidy, 2018).

Organic amendments alone, particularly maize and quinoa straw, showed moderate increases in vegetative growth, likely due to the enhancement of microbial populations and the gradual release of nutrients that support plant development (El-Hadidy, 2018).

Interestingly, the chemical fungicide Folicure, while effective in pathogen suppression, resulted in relatively lower vegetative growth metrics compared to bio-organic combinations. This outcome reflects the limitation of chemical treatments in providing growth-promoting benefits beyond pathogen control, as they do not contribute to soil health or microbial-mediated nutrient cycling (Lamichhane, *et al.* 2017; Farhaoui, *et al.* 2022 and Farhaoui, *et al.* 2024).

The infested untreated control exhibited the lowest vegetative growth, confirming the negative impact of *R. solani* infection on sugar beet development. Meanwhile, the moderate performance observed in the autoclaved soil control indicates that absence of pathogen pressure alone does not guarantee maximal growth, emphasizing the additional benefits provided by bio-organic amendments in promoting plant vigor. Overall, these results demonstrate that bio-

organic strategies integrating antagonistic fungi with organic matter represent a sustainable and effective approach to enhance sugar beet growth under pathogen pressure.

The present study demonstrated that all tested treatments significantly suppressed post-emergence damping-off, disease incidence, and severity in sugar beet under naturally infested field conditions. The untreated control recorded the highest disease levels, confirming the strong pathogen pressure in the experimental field. These findings are consistent with previous studies showing that *R. solani* and other soilborne pathogens severely reduce sugar beet emergence and growth when left unmanaged (Lamichhane, *et al.* 2017).

Chemical treatment with Folicure provided the greatest suppression of all disease parameters, which aligns with reports on the high efficacy of systemic fungicides in controlling soilborne pathogens (Lamichhane, *et al.* 2017; Farhaoui, *et al.* 2022 and Farhaoui, *et al.* 2024). However, integrated applications of *C. globosum* with organic amendments, particularly compost, also demonstrated substantial disease reduction. This suggests a synergistic effect, whereby the antagonistic fungus suppresses pathogen development while the organic amendment improves soil microbial activity and plant resilience (Al-Tawarah, *et al.* 2024).

Sole applications of organic amendments, such as rice straw, maize straw, or quinoa straw, were less effective than their combinations with *C. globosum*. This outcome supports previous findings indicating that while organic matter can suppress disease by enhancing microbial competition and improving soil properties, its efficacy is markedly increased when combined with biocontrol agents (Chen, *et al.* 2023 and Tang, *et al.* 2023). Quinoa straw alone exhibited moderate suppression, likely due to its slower decomposition and lower nutrient release compared to compost, which can limit the proliferation of beneficial microorganisms that compete with pathogens (Chaudhury, *et al.* 2023).

The results demonstrated that all tested treatments significantly influenced foliar and root growth of sugar beet under naturally infested field conditions. The untreated control consistently exhibited the lowest growth parameters, highlighting the detrimental effect of soilborne pathogens on plant development. This finding aligns with previous studies reporting severe reductions in sugar beet growth due to *R. solani* and related soilborne pathogens (Farhaoui, *et al.* 2022 and Farhaoui, *et al.* 2024).

Integrated applications of *C. globosum* with compost produced the highest improvements in both foliar and root traits, including foliar length (61.67 cm), foliar fresh weight (1235.00 g), root length (57.33 cm), and root fresh weight (1852.33 g). This indicates a synergistic effect between the biocontrol fungus and organic amendments, likely resulting from enhanced pathogen suppression and improved nutrient availability. Such synergism has been reported in several studies, where the combination of antagonistic fungi with organic matter promoted plant growth and mitigated disease pressure more effectively than either treatment alone (Yu, *et al.* 2025 and El-Hadidy, 2018).

Combinations of *C. globosum* with quinoa, rice, or maize straw also enhanced growth compared to controls, but the magnitude of improvement was lower than that observed with compost. The superior performance of compost may be attributed to its higher nutrient content and faster decomposition, which promote beneficial microbial activity and increase plant nutrient uptake (Tang, *et al.* 2023 and

Wang, *et al.* 2020). *C. globosum* alone improved growth compared to the infested control, but the lack of organic substrates likely limited its full potential, emphasizing the importance of combining biocontrol agents with suitable organic amendments (Lewaa and Zakaria 2023).

Overall, integrating *C. globosum* with compost proved to be the most effective strategy for improving both foliar and root growth traits of sugar beet under natural field infection, highlighting its potential for sustainable disease management and yield enhancement.

The findings indicated that all treatments assessed significantly influenced soil microbial populations in comparison to the untreated control. This suggests that both biocontrol agents and organic amendments can effectively alter soil microbiota in conditions of natural infestation. Soils treated with *C. globosum*, whether applied alone or alongside organic amendments, exhibited the highest fungal counts, ranging from  $40.30$  to  $42.50 \times 10^6$  CFU g<sup>-1</sup>. These results support the idea that *C. globosum* can establish itself in the rhizosphere and may outcompete pathogenic fungi, aligning with previous studies (Amin *et al.*, 2016 and El-Hadidy, 2018).

The combination of *C. globosum* with compost achieved the highest bacterial population ( $111.00 \times 10^5$  CFU g<sup>-1</sup>), followed by quinoa and rice straw combinations. This enhancement likely results from increased nutrient availability and improved soil structure provided by organic amendments, which stimulate beneficial bacterial growth (Jusoh, *et al.* 2013).

Actinomycetes, important for disease suppression and organic matter decomposition, were also significantly increased by integrated treatments, with counts peaking in soils treated with *C. globosum* + compost ( $15.57 \times 10^5$  CFU g<sup>-1</sup>) and *C. globosum* + quinoa straw ( $15.43 \times 10^5$  CFU g<sup>-1</sup>). These results align with studies demonstrating that combined biocontrol and organic amendments promote diverse microbial communities that enhance soil suppressiveness against pathogens (Silva, *et al.* 2022).

In contrast, the chemical fungicide Folicure drastically reduced microbial populations across all groups, with fungal, bacterial, and actinomycete counts falling to  $10.33 \times 10^6$  CFU g<sup>-1</sup>,  $20.13 \times 10^5$  CFU g<sup>-1</sup>, and  $4.82 \times 10^5$  CFU g<sup>-1</sup>, respectively. This suppression of beneficial microbiota is consistent with prior findings that fungicides can negatively impact non-target soil microorganisms, potentially reducing soil resilience and long-term fertility (Amin *et al.*, 2016 and El-Hadidy, 2018).

Organic amendments applied alone moderately increased microbial populations, with compost outperforming rice, maize, and quinoa straw in most microbial groups. These findings suggest that while plant residues provide nutrients and improve soil structure, the addition of an active biocontrol fungus amplifies microbial growth and diversity, likely enhancing disease suppression and soil health (Ghorbani, *et al.* 2008 and Bonanomi, *et al.* 2018).

Overall, the integration of *C. globosum* with compost was the most effective approach to boost soil microbial abundance and diversity, supporting sustainable soil management and pathogen suppression strategies, while chemical fungicide treatments, despite disease control, negatively affected beneficial soil microbiota (Saha, *et al.* 2016).

The differences observed among rice straw, maize straw, and compost amendments can largely be attributed to their chemical composition and rate of decomposition. Compost, being a pre-decomposed organic amendment, is

generally matured under controlled conditions and meets specific maturity criteria (such as reduced C/N ratio and stabilized organic matter), which ensures rapid mineralization and a steady supply of plant-available nutrients (e.g., nitrogen, phosphorus) upon soil incorporation (Bernal, *et al.* 2009).

In contrast, crop residues like rice and maize straw typically exhibit high C/N ratios and rigid lignocellulosic structures, which slow microbial decomposition and can temporarily immobilize nitrogen in the soil during the initial phases of breakdown (Yang, *et al.* 2023). However, as these residues gradually decompose, they significantly enhance soil organic matter content, stimulate microbial diversity, and boost enzyme activities—particularly ligninolytic and oxidative enzymes that contribute to pathogen suppression and soil health (Yang, *et al.* 2022).

Specifically, the study by Yang *et al.* (2022) demonstrated that straw returning substantially increased soil fertility parameters and enzyme activities (e.g., sucrase, urease, cellulase) while also reshaping soil bacterial and fungal communities in maize fields. Similarly, the more recent analysis confirmed that straw return enhances nutrient release, microbial counts, and overall soil health compared to untreated controls (Yang, *et al.* 2023).

The study revealed significant variations in peroxidase (POD) and polyphenol oxidase (PPO) activities across different treatments. Notably, the highest POD activity was observed in soils amended with compost combined with *C. globosum* ( $3804.40$  U mg<sup>-1</sup> protein), followed by quinoa straw ( $3018.23$  U mg<sup>-1</sup> protein), and rice straw combined with *C. globosum* ( $2447.85$  U mg<sup>-1</sup> protein). Similarly, PPO activity peaked in the compost and *C. globosum* treatment ( $815.14$  U mg<sup>-1</sup> protein), with quinoa straw ( $221.88$  U mg<sup>-1</sup> protein) and rice straw ( $313.21$  U mg<sup>-1</sup> protein) also showing substantial increases.

These findings align with recent studies highlighting the role of *C. globosum* in enhancing plant defense mechanisms. For instance, Elshahawy and Khattab (2022) reported increased POD and PPO activities in maize plants treated with *C. globosum*, suggesting its potential in boosting plant resistance to pathogens. Furthermore, the combination of organic amendments with *C. globosum* has been shown to synergistically enhance these enzyme activities, thereby improving plant health and disease resistance (Lewaa and Zakaria 2023). In conclusion, the integration of *C. globosum* with organic amendments proved to be the most effective strategy in enhancing POD and PPO activities, thereby bolstering the sugar beet plants' defense mechanisms. These results underscore the importance of adopting sustainable agricultural practices that not only control diseases but also promote plant resilience through natural means.

The synergistic effect between organic amendments and *Chaetomium* is particularly noteworthy. *C. globosum* is well-documented for producing cell wall-degrading enzymes such as cellulases and chitinases, as well as a variety of secondary metabolites with strong antifungal properties (Elshahawy and Khattab 2022). The presence of decomposing straw and compost likely facilitated its colonization and enzymatic activity, leading to improved suppression of *R. solani*. Furthermore, the biochemical assays confirmed that peroxidase (POD) and polyphenol oxidase (PPO) activities were significantly enhanced under straw- and compost-based treatments with *Chaetomium*, which are directly linked to the reinforcement of host defense barriers and oxidative stress responses (Lewaa and Zakaria 2023).

Overall, the integration of organic amendments with antagonistic fungi such as *Chaetomium globosum* functions

through a dual mechanism: (i) improving soil conditions by altering the microbial community structure and nutrient profile via the decomposition of organic residues, and (ii) stimulating host plant defense responses through the induction of defense-related enzymes. Consequently, these mechanisms translated into enhanced plant growth, reduced disease incidence, and suppression of root rot severity under both greenhouse and field conditions. Such integrated approaches highlight the potential of eco-friendly strategies as sustainable alternatives to chemical fungicides in the management of soil-borne pathogens (El-Tarabily, 2004; Haque and Parvin, 2021; Rehrou *et al.*, 2024 and Alqahtani, 2025).

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## المكافحة الحيوية لمرض تعفن الجذور الناتج عن فطر *Rhizoctonia solani* في بنجر السكر باستخدام المدخلات العضوية وفطر *Chaetomium globosum*: تقييم في الصوبة والحقل.

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### الملخص

الريزوكتونيا سولاني هو مرض يسبب تعفن الجذور ونبول بادرات البنجر السكري، ويشكل عائقاً رئيسياً أمام إنتاجه في مصر، إذ يسبب خسائر كبيرة في المحصول وانخفاضاً في الإنتاجية. في هذه الدراسة، تم الحصول على سبعة وعشرين عزلة من فطر *Rhizoctonia solani* من نباتات بنجر سكري مصابة بشكل طبيعي تم جمعها من محافظات الجيزة والشرقية والقليوبية. كشفت اختبارات إمراضية أجريت في ظروف البيت الزجاجي عن تباين كبير في شدة الفوعة، حيث تراوح نبول ما بعد الإنبات بين ١٠٪ و ٣٥٪. تم اختيار العزلة الأكثر عدوانية، RHI (٣٥٪)، لمزيد من التقييم. أظهرت التجارب في أصص داخل البيت الزجاجي أن دمج فطر *Chaetomium globosum* مع السماد العضوي قلل بشكل كبير من الإصابة بالمرض. انخفض نبول ما بعد الإنبات إلى ٥،٥٣٪ مقارنة بـ ٤٥،٣٧٪ في النباتات غير المعالجة، بينما انخفضت شدة المرض إلى ٨،٣٧٪ مقابل ٦٥،٣٣٪. أكدت التجارب الحقلية تحت ظروف العدوى الطبيعية هذه النتائج، حيث تم تقليل الإصابة بالمرض إلى ١٠،٢٪ في القطع المعالجة مقارنة بـ ٦٢،٧٪ في القطع الضابطة. تم تعزيز معايير النمو الخضري بشكل ملحوظ في النباتات المعالجة. في تجارب البيت الزجاجي، سجلت النباتات التي تلقت المعالجة المتكاملة ١٨،٦٧ ورقة لكل نبات، و ٣١،٨٣ سم في الارتفاع، و ٢٥،٣٠ جم في الوزن الطازج، و ١،٦٣ جم في الوزن الجاف، متفوقة بشكل كبير على النباتات الضابطة (١٢،٣٣ ورقة، ١٨،٢٠ سم، ١٢،٢٣ جم، و ٠،٨٧ جم، على التوالي). أظهرت النتائج الحقلية تحسينات مماثلة في المحصول ووزن الأصيل. كشفت الفحوصات الكيميائية الحيوية عن تحفيز قوي للإنزيمات المرتبطة بالدفاع في النباتات المعالجة. بلغت نشاط إنزيم البيروكسيداز ذروته عند ٠،٧٣١  $\Delta OD \cdot min^{-1}$  بعد ١٥٠ ثانية في المعالجة بالسماد العضوي + فطر *C. globosum*، مقارنة بـ ٠،٢١٤  $\Delta OD \cdot min^{-1}$  في النباتات الضابطة. ووصل نشاط إنزيم البولي فينول أوكسيداز إلى ٠،٤٨٧  $\Delta OD \cdot min^{-1}$  مقابل ٠،١٣١  $\Delta OD \cdot min^{-1}$  في النباتات غير المعالجة. أشار التحليل الميكروبيولوجي للتربة إلى زيادة أعداد الميكروبات النافعة في التربة المعالجة. ارتفعت أعداد الفطريات إلى ٤٣  $\times 10^3$  CFU g<sup>-1</sup> وأعداد البكتيريا إلى ١١٠  $\times 10^5$  CFU g<sup>-1</sup>، والأكتينوميسيتات إلى ١٦  $\times 10^4$  CFU g<sup>-1</sup>، مقارنة بـ ٤٠  $\times 10^3$  CFU g<sup>-1</sup>، و ١٠  $\times 10^5$  CFU g<sup>-1</sup>، و ١٠٤  $\times 10^4$  CFU g<sup>-1</sup> في التربة الضابطة، على التوالي. توضح هذه النتائج أن دمج السماد العضوي مع الفطر الحيوي *C. globosum* يثبط بفعالية فطر *R. solani*. يوفر هذا النهج الصديق للبيئة بديلاً واعداً للمبيدات الفطرية الكيميائية لزراعة البنجر السكري المستدامة.