The Pharma Innovation Journal 2023; 12(1): 3089-3095

www.ThePharmaJournal.com

The Pharma Innovation

**ISSN (E): 2277-7695**
**ISSN (P): 2349-8242**
**NAAS Rating: 5.23**
TPI 2023; 12(1): 3089-3095

www.thepharmajournal.com
Received: 22-11-2022
Accepted: 24-12-2022

**D Mahato**
Department of Plant Pathology, Institute of Agricultural Sciences, Siksha O Anusandhan (Deemed to be University), Bhubaneswar, Odisha, India

**B Khamari**
Department of Plant Pathology, Institute of Agricultural Sciences, Siksha O Anusandhan (Deemed to be University), Bhubaneswar, Odisha, India

**A Patel**
Department of Plant Pathology, Institute of Agricultural Sciences, Siksha O Anusandhan (Deemed to be University), Bhubaneswar, Odisha, India

**Corresponding Author:**
**D Mahato**
Department of Plant Pathology, Institute of Agricultural Sciences, Siksha O Anusandhan (Deemed to be University), Bhubaneswar, Odisha, India

# Effect of autoclaved paddy straw-based oyster spent mushroom substrate on elicitors, mineral content, growth stimulation and root rot suppression of sesame

**D Mahato, B Khamari and A Patel**

**Abstract**
A substantial difficulty has been presented by the recent decade's fast growth of the edible mushroom business worldwide, which has led to a large generation of SMS. Environmental issues including soil contamination, air pollution, and water pollution can result from improper management. With a focus on "circular economy," SMSs have shown tremendous promise in a variety of uses, such as a plant protectant. In this experiment the protective effect of Spent Oyster Mushroom Substrate (SMS) aqueous extracts were examined *in vitro* for their ability to combat *Macrophomina phaseolina causing* root rot of sesame. Employing methodological approaches, the physiochemical characteristics and elicitors in the treatments were evaluated which revealed differences in the values of elicitors: carbohydrate polymers (13.54-16.53%), glycoproteins (0.32-0.52%) and lipid molecules (0.31-12.64 g) present between the treatments. The non-autoclaved aqueous extract of SMS recorded better mycelial inhibition of *Macrophomina phaseolina* at different concentration (1%, 5%, 10%, 15% and 20%) as compared to autoclaved SMS with maximum mycelia inhibition at 20% in poison food technique. In pot culture experiment, spent mushroom substrate to soil suppressed the disease severity to an appreciable limit (64.63%) and promoted plant growth parameters recording plant heights (43.40 cm), number of leaves (11.3), number of flowers (6.3), number of capsules (8.0), weight of capsule (12.55), fresh weight (3.04 g) and dry weight (1.31 g) of plant. These findings suggest that water-soluble and heat-stable compounds in SMS enhance the state of systemic acquired resistance and suppress root rot of sesame and augment plant growth. Thus, the use of SMS for disease control may offer a new technology for the recycling and management of waste from mushroom cultivation.

**Keywords:** *Macrophomina phaseolina*, mushroom, *Pleurotus florida*, root rot, spent mushroom substrate, circular economy, sustainable agriculture

## Introduction
Mushroom which has a high nutritional, therapeutic properties, ecological importance, emerged as a popular food due to the rising trend of healthy eating (Valverde *et al*., 2015) [46]. Despite the evident benefits of mushrooms, the exponential increase and economic significance of mushroom which push its production globally and predicted to touch 20.84 million tons by the year 2026 (Atallah *et al*., 2021) [3] has generated a high volume of spent mushroom substrate (SMS). Rapid increase of mushroom industry results into excess piling of spent mushroom substrate (SMS) or spent mushroom compost (SMC) with annual production of more than 60 million tons SMS (Atallah *et al*., 2021) [3]. These piles treated as waste and are disposed in the form of landfilling, incineration or open burning, composting with animal manure without proper utilization. The improper dispose may lead to soil contamination, air and water pollution (Jiang *et al*., 2017; Lam *et al*., 2019) [16, 25]. There should be an efficient recycling and utilization of these SMS to support circular economy, promoting environmental conservation (Pérez-Chávez *et al*., 2019; Mahari *et al*., 2020) [37, 29], because it has an immense potential as substrate for new cultivation cycle of mushroom, bio-fertilizer, soil amendment, animal feed, renewable energy production and pollution bioremediation (Leong *et al*., 2022) [26]. It mainly composed of residual fungal mycelium, disintegrated lignocellulosic substrate biomass, amendments, nutrients and high level of organic matter as well as enzymes (Najafi *et al*., 2019) [31] which greatly depends on location and type of mushroom (Leong *et al*., 2022) [26]. Being an excellent source of carbon, nitrogen and other essential elements, spent mushroom substrate can be used as very good soil conditioner (Prasad and Srinivasaraghavan 2019) [39] as it enriches soil health, augments plant growth and enhances microbial population (Singh *et al*., 2018) [44].

In plant-fungal interactions, the mycelia of fungal pathogens release proteins and carbohydrates (elicitors) that trigger plant defence mechanisms (Shibuya and Minami, 2001) [43] These chitin-based compounds (SMS) are rich sources of elicitors for plant defence, allowing plants to withstand or tolerate a wide range of diseases (Parada *et al*., 2011) [36]. The complicated interactions between abiotic factors, microbial populations and host result in suppression of soil-borne pathogens exploiting SMS. It is linked to antibiosis, microbial production of lytic enzymes, parasitism, changes in nutritional availability and host-mediated development of resistance. It reduces severity of soil-borne diseases in vegetable crops by promoting a population of antagonistic microorganisms (Prasad and Srinivasaraghavan, 2019) [39]. Agaricus-Pleurotus mixed SMS showed the higher suppressiveness (50%) against *Pythium irregulare* than sole Agaricus (38%) or Pleurotus (15%) supplemented media. Spent mushroom substrate also suppressed foliar disease incidence such as powdery mildew and angular leaf spot of cucumber (Parada *et al*., 2012) [35], sooty spot of cabbage, Phytopthora blight of chilli (Kang, 2017) [17]. Water extract of SMS suppressed the development of lesion and inhibited conidial germination of *Pyricularia oryzae* (Ishihara *et al*., 2018, 2019) [14].

Root rot is a devastating soil borne disease, causing huge economic loss in sesame (Khamari *et al*., 2018) [20]. The broad host range and wider adaptability of the pathogen associated, makes the disease management more challenging (Gulya *et al*., 2002; Ransingh *et al*., 2021) [12, 40]. Due to ease of use and capacity to provide quick treatment, chemical fungicides are farmers' primary choice for disease management but it is hazardous to human health and environment. The biological strategy of treating plant diseases using composts and biological control agents is the best alternative for it (Khamari and Hasmi, 2019; Khamariet *et al*., 2021, 2022) [19, 18]. Looking into these literatures, it was assumed that SMS can be a good alternative to ameliorate the deadly disease of sesame. In this connection, the current experiment was designed with an objective to assess the effectiveness of SMS for root rot diseases suppression in sesame *in vitro.*

## Methods

### Disease specimen and associated pathogen

Diseased sesame plants showing characteristic symptoms of root rot /stem rot (Khamari *et al*., 2016) [21] were collected from the farmers field, Bargarh district, Odisha, India (latitude of 21.2550° N and longitude of 83.5070° E) and the causal pathogen was isolated using standard protocol and identified as *Macrophomina phaseolina.*

### Molecular identification of *Macrophomina phaseolina*

The fungal DNA was isolated and amplified using 18s rRNA specific primer i.e. ITS 1 (5'-TCCGTAGGTGAACCTGCGG-3') and ITS 4 (5'-TCCTCCGCTTATTGATATGC-3') using Veriti® 96 well Thermal Cycler with following conditions, initial denaturation at 95 °C for 5 mins followed by 39 cycles of denaturation 30 sec at 95 °C, 45 sec annealing and 1 min elongation at 72 °C, with a final extension of 7 min at 72 °C. The amplicons were resolved in 1.5% agarose gel with ethidium bromide stain using 0.5x trisacetate-EDTA (TAE) buffer and documented amplification. Bi-directional DNA sequencing reaction of PCR amplicon was carried out using BDT v3.1 Cycle sequencing kit on ABI 3500Dx Genetic Analyzer. For identification, the PCR products were eluted using QIA quick gel extraction kit and sequenced using ABI 3730 XL sequencer. The amplified PCR products were sequenced by Sanger's dideoxy sequencing method (Sanger *et al*., 1977) [42]. Further, both the forward and reverse sequences were reviewed and aligned using BioEdit (version 7.2.5) to generate consensus sequences. It was further analysed for similarity index through BLASTn program of NCBI (National Center for Biotechnology the Information). The sequence thus obtained was submitted to NCBI Genbank for generating accession number followed by phylogenetic and molecular evolutionary analysis. The most closely matched sequences were downloaded from BLAST and aligned using MUSCLE of MEGA (Molecular Evolutionary Genetics Analysis) software. The phylogenetic tree was constructed through neighbor joining method (Saitou and Nei, 1987) [41] with 1000 bootstrap value (Felsenstein, 1985) [8] in MEGA11 software.

### Preparation of treatments

#### Spent mushroom substrate

Oyster mushroom (*Pleurotus florida*) grown in sterilized chopped straw in a polythene bag of 5kg capacity, supplemented with gram flour and harvested in 3 flushes at 17$^{th}$, 21$^{st}$ and 24$^{th}$ day at Mushroom unit, SOADU, Bhubaneswar, Odisha, India. After a complete harvest, the spent mushroom substrates (SMS) were chopped into small pieces and stored at 4 ºC for later use. For *in vitro* experiments, the autoclaved water extract of SMS (AWESMS) and non-autoclaved water extract of SMS (NWESMS) was prepared by following the standard protocol described by Parada *et al*., (2011) [36] and Adedeji *et al*., (2016) [1] with some modifications and stored for future use.

### Evaluation of the Physiochemical parameters and elicitors

Mineral composition of the treatments and pH was determined using atomic spectrophotometer and pH meter at the Soil Science Department, SOA University Bhubaneswar. The elicitors were determined according to the slandered procedure described by Okere and Ataga (2018) [33] using a spectronic 21D digital spectrophotometer. The presence of glycoprotein was determined at a wavelength of 495nm and calculated using the given formula

$$\frac{\text{Absorbance of sample x gradient factor x dilution factor if any}}{\text{Weight of sample taken for protein glycosylation}} \times 100$$

The total soluble carbohydrate was determined at a wavelength of 620nm and then estimated using the standard curve of Glucose (Okere and Ataga, 2018) [33]. The lipid extraction was carried out by soaking two gram each of the sample in 50 ml of distilled water over night, further the solution was sieved using muslin cloth and mixed with 50 ml of ethanol and was transferred into a separating funnel positioned with retort stand followed by addition of 50 ml of petroleum ether into the separating funnel and allowing them to stand for about five minutes. The residue containing the lipids was transferred to a pre-dried and weighed 50 ml beaker. The pre- dried and weighed beaker with the content were oven dried between (40-700 C) for a few minutes (W1). The beaker and content were allowed to cool and re-weighed as W2. The quantity of lipid (oil) extracted was calculated as follows: W2-W1 (Okere and Ataga, 2018) [33].

**Antifungal efficacy of WESMS and AWESMS**

Using the technique of poisoned food, the effectiveness of SMS on *M. phaseolina in vitro* radial growth was examined. To acquire concentrations of 1% (0.01 g/ml), 5% (0.05 g/ml), 10% (0.1 g/ml), or 15% (0.15 g/ml) of the prepared SMS, 1g, 5 g, 10 g, 15 g, or 20 g of each were first suspended in 100ml of sterile DW. These suspensions were then further agitated by vigorously shaking to ensure even particle distribution. Ten millilitres of warmed, melted PDA were blended with two portions of one mililiter each of AWESMS and WESMS before being gently swirled and put on petridish. A 5 mm diameter mycelial disc was placed aseptically in the centre of a petri dish and incubated at 28 °C. A control treatment was combined with a cooled PDA that contained 1 milliliter of sterilised distilled water without SMS. Each treatment replicated 4 times. The radial growth of *M. phaseolina* was recorded in different time interval and per cent inhibition was calculated using Vincent formulae (Vincent, 1947) [47].

**Protective effect of SMS and ASMS mixed in soil on root rot of sesame**

To observe the disease-protective effect of SMS and ASMS mixed in soil, sesame seedlings (Mahadev (Sec551)) were planted in pots containing mixtures of SMS or ASMS and soil (1:2, v/v) and tested for disease protection. All the treatments were challenged by inoculating with previously mass multiplied *Macrophomina phaseolina* inoculums at the rate of 4g/kg as per procedure given by Khamari *et al*., (2019) [19]. The experiment was designed in a completely-randomised design with four treatments and five replications. Number of germinated seedlings was counted on 15$^{th}$ day and germination percentage were calculated. Seedling length was also recorded and Vigour index was calculated. Disease severity was measured at 45$^{th}$ and 60$^{th}$ day using modified Cralley's system of disease measurement (Bhattacharyya *et al*., 1985) [4] scale such as 0-No lesions on plants, 1-Number of slightly appeared lesions on plants, 2-Number of heavily appeared lesions on plants, 3-Weak plants/root rot disease of plants, 4-Dead plants. The percent disease severity index was calculated by the formulae below.

$$DSI\% = \frac{\sum(\text{Grade X Number of plants in that grade})}{\text{Total number of accessed plants}} X100$$

Different plant growth traits and yield attributing characters such as plant height, number of leaves, number of flowers per plant, number of capsules per plant, weight of capsule, fresh and dry weight of plant biomass were recorded during maturity.

**Statistical analysis**

All the experiments were designed using completely randomized design. Data obtained from the experiments were analysed by analysis of variance (ANOVA) using SPSS Var-27 software. The means were compared with 95% level of significance ($p$<0.05).

**Results**

**Molecular identification of root rot pathogen**

The consensus sequence obtained after processing with BioEdit was analysed for similarity index on NCBI database, i.e., BLASTn and phylogenetic tree was constructed verify a correlation between the host species, the geographic area and the pathogen. The sequence of ITS region showed phylogenetic link with *Macrophomina phaseolina* by 100% homology (Figure 1). The sequence was submitted to NCBI GenBank database and the accession number OP647417 was generated. The molecular studies and analysis exhibited good agreement with that of morphological studies and confirm the identity of the fungal species under evaluation.

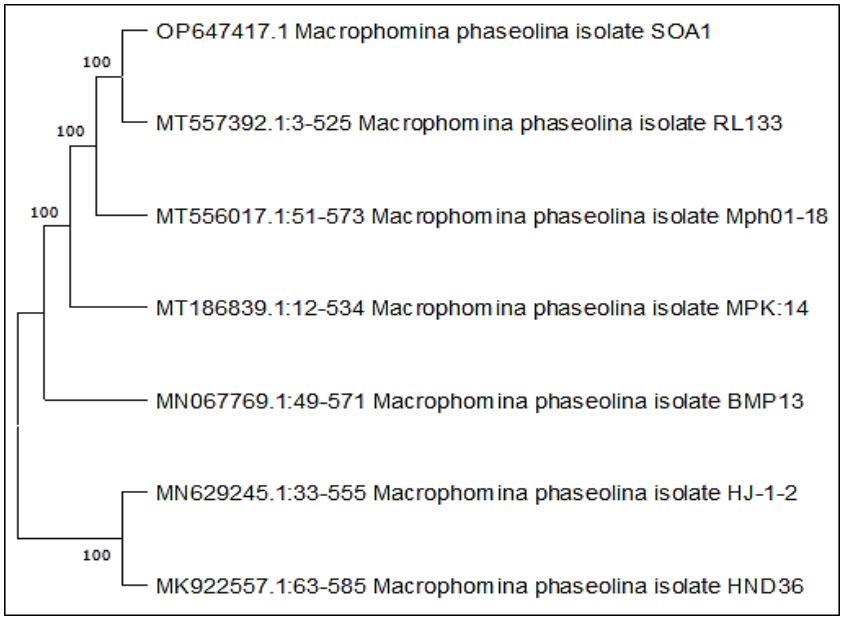


**Fig 1:** Phylogenetic tree of *Macrophomina phaseolina* based on ITS sequence. (The red mark represent the sequence generated from this study)

**Physicochemical characteristic analysis of treatments**

The physiochemical characteristics of NWESMS and AWESMS, involving pH, EC, N, P and K content, were shown in Table 1. The pH of NWESMS was slightly higher (7.20) than AWESMS (7.17). NWESMS had 0.31 mg less nitrogen than AWESMS and had 39.93 and 5.18 mg higher concentrations of phosphorus and potassium than AWESMS.

**Table 1:** Physiochemical characteristics of Spent mushroom substrate

| Parameters | NWESMS | AWESMS |
|---|---|---|
| pH | 7.20 | 7.17 |
| Nitrogen (mg/100g) | 3.16 | 3.47 |
| Phosphorus (mg/100g) | 210.56 | 170.63 |
| Potassium (mg/100g) | 17.55 | 12.37 |

NWESMS = Non-autoclaved water extract spent mushroom substrate. AWESMS = Autoclaved water extract spent mushroom substrate

### Evaluation of elicitors

Three elicitors were recorded in spent mushroom substrate such as carbohydrate polymers, glycoproteins and lipid molecules. NWESMS included 12.33g less lipid molecules than AWESMS but contained 2.99% and 0.2% more carbohydrate and glycoprotein molecules than AWESMS, respectively (Table 2).

**Table 2:** Evaluation of the elicitors in the autoclaved and unautoclaved water extract SMS

| Treatment | Carbohydrate polymers (%) | Glycoproteins (%) | Lipid molecules (g) |
|---|---|---|---|
| NWESMS | 16.53$^{a}$ | 0.52$^{a}$ | 0.31$^{b}$ |
| AWESMS | 13.54$^{b}$ | 0.32$^{b}$ | 12.64$^{a}$ |
| LSD (P=0.05) | 1.61 | 0.17 | 2.60 |

### Efficacy of aqueous SMS extract on mycelia inhibition of *M. phaseolina in vitro*

Non-autoclaved water extract of spent mushroom substrate (NWESMS) found superior over autoclaved water extract of spent mushroom substrate (AWESMS) in mycelial inhibition (Table 3). Maximum mycelial inhibition was recorded at 20% conc. (54.95%) followed by 15% (45.05) and 10% (42.34) in NWESMS. Similarly, in AWESMS 20% registered maximum inhibition followed by 15% and 10% recording 45.95%, 44.14% and 32.43% respectively at 3 days after inoculation (DAI). Similar trend was observed at 5DAI recording maximum inhibition (69.07%) in 20% concentration of NWESMS. It was observed that, mycelial inhibition increases with increase in the concentration of SMS. Comparing autoclaved spent mushroom substrate (AWESMS) to non-autoclaved SMS, the NWESMS exhibits a significantly stronger inhibitory action of mycelium development. The highest inhibition percentage was recorded at 20% concentration.

**Table 3:** Effects of autoclaved and non-autoclaved Spent Mushroom Substrate on *Macrophomina phaseolina* growth

| Inhibition percentage (%) (average) | | | | |
|---|---|---|---|---|
| Concentration (g/ml) | 3 DAI | | 5 DAI | |
| | NWESMS* | AWESMS* | NWESMS | AWESMS |
| 0.01 | 20.72 | 10.81 | 34.76 | 13.39 |
| 0.05 | 31.53 | 12.61 | 57.26 | 15.01 |
| 0.1 | 42.34 | 32.43 | 64.00 | 38.13 |
| 0.15 | 45.05 | 44.14 | 65.69 | 46.57 |
| 0.2 | 54.95 | 45.95 | 69.07 | 60.07 |
| Control | 0.00 | 0.00 | 0.00 | 0.00 |
| LSD(P=0.05) | 0.433 | 0.541 | 0.609 | 0.624 |

***AWESMS:** Autoclaved water extract SMS.
***WESMS:** Non-autoclaved water extract SMS.

### Pot culture experiment

### Efficacies of SMS on root rot disease severity and growth augmentation

The disease severity was observed more at 60 days after sowing (DAS) ranging from 25.30% to 71.54% as compared to 45 DAS (22.63% to 57.53%). All the treatments significantly suppressed the disease incidence at 45 DAS and 60 DAS. Soil mixed with SMS reduces maximum disease severity over control at 45 DAS (60.66%) and 60 DAS (64.63%) whereas soil without SMS (Control) showed least efficacy against root rot disease (Table 4). The disease severity was recorded maximum in control at 45 DAS (57.53%) and 60 DAS (71.54%) (Table 4). Plant growth parameters were significantly ($p$<0.05) increased upon SMS incorporation. Maximum plant height (37.06cm), number of leaves per plant (12.93), number of capsules per plant (9.25), fresh (3.04gm) and dry weight of plant biomass (3.29gm) was observed in SMS treatment followed by ASMS. The lowest plant growth parameters were observed in control (Table 4).

**Table 4:** Efficacy of SMS in suppression of root rot of sesame

| Treatment | Disease severity (%) | Efficacy (%) | Disease severity (%) | Efficacy (%) | Plant height (in cm) | No. of leaves per plant | No. of capsules per plant | Weight of plant biomass | |
|---|---|---|---|---|---|---|---|---|---|
| | 45 DAS | | 60 DAS | | | | | Fresh weight (in gm) | Dry weight (in gm) |
| SMS | 22.63$^{b}$ | 60.66 | 25.30$^{b}$ | 64.63 | 37.06$^{a}$ | 12.93$^{a}$ | 9.25$^{a}$ | 3.04$^{a}$ | 3.29$^{a}$ |
| ASMS | 27.9$^{b}$ | 51.50 | 30.70$^{b}$ | 57.09 | 31.24$^{ab}$ | 11.8$^{a}$ | 7.05$^{ab}$ | 2.38$^{a}$ | 2.38$^{a}$ |
| Soil (Control) | 57.53$^{a}$ | - | 71.54$^{a}$ | - | 23.75$^{b}$ | 6.00$^{b}$ | 5.25$^{b}$ | 1.3$^{b}$ | 1.3$^{b}$ |
| LSD (P=0.05) | 12.077 | | 6.547 | | 7.894 | 3.972 | 2.509 | 0.655 | 0.93 |

### Discussion

Spent mushroom substrate, an important by product of mushroom industry, consisted of residual fungal mycelium, a wide variety of disintegrated lignocellulosic biomass, amendments, nutrients, and enzymes (Najafi *et al*., 2019) [31], has an immense potential in suppression of pathogenic microorganisms while providing essential soluble nutrients for plant assimilation. Being an excellent source of carbon, nitrogen, and other essential protein-rich component, it contributes significant benefaction to soil health which subsequently upgrades crop productivity (Singh *et al*., 2018) [44]. Following analysis of the physiochemical characteristics, it was discovered in the current study that there are few differences between the NWESMS and AWESMS in terms of nitrogen content. This finding is consistent with the findings of (Okere and Ataga, 2018) [33] and suggests that nitrogen may be both heat and water soluble. Additionally, the results showed that autoclaving had an impact on the amounts and availability of phosphorus in the water-extract spent mushroom substrate, indicating that they are not heat-stable. The lowest amounts of potassium were found in the water extract from both SMS, indicating that potassium is less water soluble but more heat stable.

In plant-fungal interactions, carbohydrate and protein elicitors that induce defense mechanism in plants are released from the mycelia of fungal pathogens (Shibuya and Minami, 2001) [43] which are recognize by the plants to develop an enhanced resistance. This type of induced resistance is called systemic acquired resistance (SAR) (Durrant and Dong, 2004; Conrath, 2006) [7, 6]. Several studies have reported that the mycelia of mushrooms that are consist of fragments of glucan and chitin may serve as elicitors for inducing SAR and thus application of SMS to plants may be useful for the control of plant diseases. In the present study evaluation of the different elicitors in the spent mushroom substrate revealed that unautoclaved water extract SMS contained 2.99% and 0.20% higher carbohydrate and glycoprotein molecules but 12.33g less lipid molecules than autoclaved water extract SMS respectively.

The suppression of soil-borne pathogens derived from agro-industrial wastes i.e. spent mushroom substrates (Noble and Coventry, 2005) [32] are reported by several studies which revealed that the suppressiveness has been attributed to complex interactions between abiotic characteristics and/or microbial populations and plants, involving microbial competition for nutrient substrates and ecological niches, antibiosis, microbial production of lytic enzymes, parasitism, changes in nutrient availability and host-mediated induction of resistance (Lorito *et al*., 1993; Borrero *et al*., 2006; Perez-Piqueres *et al*., 2006) [28, 5, 38]. It also induces defence responses in plants (Parada *et al*., 2012; Prasad and Srinivasaraghavan 2019) [35, 39]. It is also reported that SMS induces resistance by accumulation of phytoalexin, PR genes, production of phenolic compounds. Accumulation of transcripts of pathogenesis-related genes (CaBPR1, CaBGLU, CaPR-4 and CaPR10) and an increase in the salicylic acid (SA) content suppressed Phytophthora blight of pepper upon *L. edodes* water extract treatment (Kang, 2017) [17].

There are a number of reports of reduction of diseases using SMS. Water extract of SMS suppressed lesion development and conidial germination of *Pyricularia oryzae* (Arase *et al*., 2013; Ishihara *et al*., 2018, 2019) [2, 14]. Spraying aquous SMS extract of *Lyophyllum decastes* (fried chicken mushroom) suppressed the blast lesion development in rice (Arase *et al*., 2013) [2]. Spraying of water extracts of *L. decastes* and *P. eryngii* SMSs on the leaves or mixing autoclaved SMS reduces powdery mildew and angular leaf spot but not effective against Corynespora leaf spot and scab (Parada *et al*., 2012) [35]. In the present study, water extract of oyster mushroom substrate reduces radial growth of *Macrophomina phaseolina*, causing root rot of sesame and promoted seed germination. Non-autoclaved water extract SMS registered more disease suppression as compared to autoclaved water extract of SMS. The severity of root rot disease was also significantly suppressed due to SMS application in pot culture experiment. The mechanisms behind suppressive effects of SMS are due to antimicrobial compounds and volatiles present in it (Osaki-Oka, 2018) [34]. SMS contains various volatile compounds such as skatole, 2,4-di-tert-butylphenol, g-dodecalactone, butyric acid, guaiacol, 6-amyl-2-pyrone and 1-octen-3-ol which inhibit pathogen and suppress hyphae elongation (Fujita *et al*., 2021) [9]. The extract of *L. edodes* SMSs contains three antimicrobial phenolic compounds (Ishihara *et al*., 2018) [15]. Si is an important element present in rice plant which increases with crop growth and plays important role in host resistance. Paddy straw SMS showed resistance against pathogen and enhance microbial interaction (Wang *et al*., 2017; Yan *et al*., 2018) [48, 49]. In the present experiment, paddy straw was used, which may the reason behind suppression of disease incidence. Application of SMS in soil not only suppressed root rot diseases incidence but also promoted seed germination and augmented sesame plant growth. Similar findings are obtained by several workers. Application of *Flammulina velutipes* substrate effectively suppresses cucumber Fusarium wilt, augmented plant growth and reduces the population of *Fusarium oxysporum* f. sp. *Cucumerinum* (Wang *et al*., 2020) [50]. When SMS applied sufficiently, correctly and appropriately, it improves soil health (Medina *et al*., 2012) [30] and plant growth (Gobbi *et al*., 2018; Li *et al*., 2020) [10, 27] by increasing number of beneficial microbes (Wang *et al*., 2020) [50]. Sönmez *et al*. (2016) [45] observed the salt content of the SMC has great contribution in germination and seedling quality. The increased organic matter in soil due to application of SMC has potentiality to partially replace chemical fertilizers (Grimm and Wosten, 2018) [11]. Application of SMS act as a bioremediation agent of infertile and contaminated soil which improve the soil structure, physico-chemical, chemical and biological properties that can increase crop yield.

## Conclusion

In conclusion, the application of SMS significantly suppresses the root rot disease severity, enhanced the seed germination and plant growth. Thus, SMS may be used as plant growth promoter and disease suppressant to support sustainable agriculture. However, advanced studies are needed to elucidate the changes in soil environment, soil nutrient cycling, soil microbial properties after the application of SMS and mechanism behind its disease suppression as well as growth promotion.

## Abbreviations

SMS: Spent mushroom substrate; DAS: days after showing; DSI: Disease severity index; DAI: Days after inoculation; NAWSMS: Non-autoclaved water extract spent mushroom substrate; AWSMS: Autoclaved water extract spent mushroom substrate; ASMS: Autoclaved spent mushroom substrate.

## Declarations

**Ethics approval and consent to participate:** Not applicable.

**Availability of data and materials:** Authors confirm availability of data supporting the findings within the manuscript itself.

**Competing of interest:** Authors declare that they have no competing of interests.

**Funding:** Not applicable.

## Author contributions

DM carried out the experimental work. BK supervised and suggested the problem. AP helped in the experimental work. All authors contributed writing draft and BK finalized the manuscript in the final form. All authors have read and approved the manuscript.

**Acknowledgements:** Not applicable.

**References**


1. Adedeji KO, Aduramigba MA. *In vitro* evaluation of spent mushroom compost on growth of *Fusarium oxysporium* f. sp. lycopersici. Advances in Plants and Agriculture Research. 2016;4(4):332-339.
2. Arase S, Kondo Y, Jaco RY, Otani H, Ueno M, Kihara JI. Suppression of rice blast disease by autoclaved water extract from the spent mushroom substrate of *Lyophyllum decastes*. Mushroom Science and Biotechnology. 2013;21(2):79-83.
3. Atallah E, Zeaiter J, Ahmad MN, Leahy JJ, Kwapinski W. Hydrothermal carbonization of spent mushroom compost waste compared against torrefaction and pyrolysis. Fuel Processing Technology. 2021;216:106-795.
4. Bhattacharyya D, Basu S, Chattapadhyay JP, Bose SK. Biocontrol of Macrophomina root-rot disease of jute by an antagonistic organism, *Aspergillus versicolor*. Plant and soil. 1985;87(3):435-438.
5. Borrero C, Ordovás J, Trillas MI, Avilés M. Tomato Fusarium wilt suppressiveness. The relationship between the organic plant growth media and their microbial communities as characterised by Biolog®. Soil Biology and Biochemistry. 2006;38(7):1631-1637.
6. Conrath U. Systemic acquired resistance. Plant signaling & behavior. 2006;1(4):179-184.
7. Durrant WE, Dong X. Systemic acquired resistance. Annual review of phytopathology. 2004;42(1):185-209.
8. Felsenstein J. Confidence limits on phylogenies: an approach using the bootstrap evolution. 1985;39(4):783-91.
9. Fujita R, Yokono M, Ube N, Okuda Y, Ushijima S, Fukushima-Sakuno E, *et al*. Suppression of Alternaria brassicicola infection by volatile compounds from spent mushroom substrates. Journal of bioscience and bioengineering. 2021;132(1):25-32.
10. Gobbi V, Nicoletto C, Zanin G, Sambo P. Specific humus systems from mushrooms culture. Applied Soil Ecology. 2018;123:709-13.
11. Grimm D, Wösten HA. Mushroom cultivation in the circular economy. Applied microbiology and biotechnology. 2018;102(18):7795-7803.
12. Gulya TJ, Krupinsky J, Draper M, Charlet LD. First report of charcoal rot (*Macrophomina phaseolina*) on sunflower in North and South Dakota. Plant disease. 2002;86(8):923.
13. Hall T, Biosciences I, Carlsbad CJ. BioEdit: an important software for molecular biology. GERF Bull Bio Sci. 2011;2(1):60-61.
14. Ishihara A, Ando K, Yoshioka A, Murata K, Kokubo Y, Morimoto N, *et al*. Induction of defense responses by extracts of spent mushroom substrates in rice. Journal of pesticide science; c2019. p. D18-063.
15. Ishihara A, Goto N, Kikkawa M, Ube N, Ushijima S, Ueno M, *et al*. Identification of antifungal compounds in the spent mushroom substrate of *Lentinula edodes*. Journal of Pesticide Science. 2018;43(2):108-113.
16. Jiang H, Zhang M, Chen J, Li S, Shao Y, Yang J, *et al*. Characteristics of bio-oil produced by the pyrolysis of mixed oil shale semi-coke and spent mushroom substrate. Fuel. 2017;200:218-224.
17. Kang DS, Min KJ, Kwak AM, Lee SY, Kang HW. Defense response and suppression of Phytophthora blight disease of pepper by water extract from spent mushroom substrate of *Lentinula edodes*. The plant pathology journal. 2017;33(3):264.
18. Khamari B, Rath A, Satapathy RR, Gogoi S. System based approach for the management of soil borne disease in Brinjal. Solanum melongena: Production, cultivation and nutrition. Nova science publishers, USA; c2021. p. 217-239.
19. Khamari B, Hasmi SK. Biointensive management of *Macrophomina phaseolina*, inciting stem and root rot of sesame. Current research and innovations in plant pathology. 2019;7:1-26.
20. Khamari B, Beura SK, Ranasingh N. Status of sesame diseases grown in different agroclimatic zones of Odisha. International Journal of Current Microbiology and Applied Sciences. 2018;7(11):945-958.
21. Khamari B, Beura SK, Ranasingh N, Dhal A. Symptomatological study of stem and root rot of sesame. Journal of Mycopathological Research. 2016;54(3):443-445.
22. Khamari B, Hasmi SK, Samal KC, Sahoo JP, Senapati AK, Ranasingh N, Beura SK, Samal T. Impact of microbial rivals and natural alterations on root decay and plant development in sesame. Indian Phytopathology. 2022;75(4):1075-1083.
23. Khamari B, Satapathy SN, Patra C. Effect of inoculum load and duration of exposure to *Macrophomina phaseolina* on disease incidence of sesame. Int. J Chem. Stud. 2019;7(1):1728-1730.
24. Kumar S, Stecher G, Tamura K. MEGA7: molecular evolutionary genetics analysis version 7.0 for bigger datasets. Molecular biology and evolution. 2016;33(7):1870-1874.
25. Lam SS, Lee XY, Nam WL, Phang XY, Liew RK, Yek PN, *et al*. Microwave vacuum pyrolysis conversion of waste mushroom substrate into biochar for use as growth medium in mushroom cultivation. Journal of Chemical Technology & Biotechnology. 2019;94(5):1406-1415.
26. Leong YK, Ma TW, Chang JS, Yang FC. Recent advances and future directions on the valorization of spent mushroom substrate (SMS): A review. Bioresource technology. 2022;344:126-157.
27. Li TH, Che PF, Zhang CR, Zhang B, Ali A, Zang LS. Recycling of spent mushroom substrate: Utilization as feed material for the larvae of the yellow mealworm Tenebrio molitor (Coleoptera: Tenebrionidae). Plos one. 2020;15(8):e023-7259.
28. Lorito M, Di Pietro A, Hayes CK, Woo SL, Harman GE. Antifungal, synergistic interaction between chitinolytic enzymes from *Trichoderma harzianum* and *Enterobacter cloacae*. Phytopathology (USA); c1993.
29. Mahari WA, Peng W, Nam WL, Yang H, Lee XY, Lee YK, *et al*. A review on valorization of oyster mushroom and waste generated in the mushroom cultivation industry. Journal of hazardous materials. 2020;400:123-156.
30. Medina E, Paredes C, Bustamante MA, Moral R, Moreno-Caselles J. Relationships between soil physico-chemical, chemical and biological properties in a soil amended with spent mushroom substrate. Geoderma. 2012;173:152-61.
31. Najafi B, Faizollahzadeh Ardabili S, Shamshirband S, Chau KW. Spent mushroom compost (SMC) as a source

for biogas production in Iran. Engineering Applications of Computational Fluid Mechanics. 2019;13(1):967-982.
32. Noble R, Coventry E. Suppression of soil-borne plant diseases with composts: a review. Biocontrol Science and Technology. 2005;15(1):3-20.
33. Okere SE, Ataga AE. Effects of autoclaving (*Pleurotus osteratus*) spent mushroom substrate water extract on the minerals, elicitors, cassava yield and the management of African cassava mosaic virus. Int. J Adv. Appl. Sci. 2021;10(1):13-9.
34. Osaki-Oka K, Kodani T, Ishihara A. Suppression of Alternaria sooty spot of cabbage by volatile compounds from spent substrates of shiitake (*Lentinula edodes*) and Bunashimeji (*Hypsizygus marmoreus*). Mushroom Sci. Biotechnol. 2018;26:28-31.
35. Parada RY, Murakami S, Shimomura N, Otani H. Suppression of fungal and bacterial diseases of cucumber plants by using the spent mushroom substrate of *Lyophyllum decastes* and *Pleurotus eryngii*. Journal of Phytopathology. 2012;160(7-8):390-339.
36. Parada RY, Murakami S, Shimomura N, Egusa M, Otani H. Autoclaved spent substrate of hatakeshimeji mushroom (*Lyophyllum decastes* Sing.) and its water extract protect cucumber from anthracnose. Crop Protection. 2011;30(4):443-450.
37. Pérez-Chávez AM, Mayer L, Albertó E. Mushroom cultivation and biogas production: A sustainable reuse of organic resources. Energy for Sustainable Development. 2019;50:50-60.
38. Pérez-Piqueres A, Edel-Hermann V, Alabouvette C, Steinberg C. Response of soil microbial communities to compost amendments. Soil Biology and Biochemistry. 2006;38(3):460-470.
39. Prasad D, Srinivasaraghavan A. Management of Soil borne Diseases of Vegetable Crops through Spent Mushroom Substrate. In the Vegetable Pathosystem. Apple Academic Press; c2019. p. 511-526.
40. Ransingh N, Khamari B, Adhikary NK. Modern Approaches for Management of Sesame Diseases. In Innovative Approaches in Diagnosis and Management of Crop Diseases. Apple Academic Press; c2021. p. 123-162.
41. Saitou N, Nei M. The neighbor-joining method: a new method for reconstructing phylogenetic trees. Molecular biology and evolution. 1987;4(4):406-425.
42. Sanger F, Nicklen S, Coulson AR. DNA sequencing with chain-terminating inhibitors. Proceedings of the national academy of sciences. 1977;74(12):5463-5467.
43. Shibuya N, Minami E. Oligosaccharide signalling for defence responses in plant. Physiological and Molecular Plant Pathology. 2001;59(5):223-233.
44. Singh UB, Malviya D, Khan W, Singh S, Karthikeyan N, Imran M, *et al*. Earthworm grazed-Trichoderma harzianum biofortified spent mushroom substrates modulate accumulation of natural antioxidants and bio-fortification of mineral nutrients in tomato. Frontiers in Plant Science. 2018;9:1017.
45. Sonmez I, Kalkan H, Demir HA. Effects of spent mushroom compost on seedling quality and nutrient contents of eggplant (*Solanum melongena*) grown in different growing media; c2016.
46. Valverde ME, Hernández-Pérez T, Paredes-López O. Edible mushrooms: improving human health and promoting quality life. International journal of microbiology; c2015.
47. Vincent JM. The esters of 4-hydroxybenzoic acid and related compounds. Part I. Methods for the study of their fungistatic properties. Journal of the Society of Chemical Industry. 1947;66(5):149-55.
48. Wang M, Gao L, Dong S, Sun Y, Shen Q, Guo S. Role of silicon on plant-pathogen interactions. Frontiers in Plant Science. 2017;8:701.
49. Yan GC, Nikolic M, Ye MJ, Xiao ZX, Liang YC. Silicon acquisition and accumulation in plant and its significance for agriculture. Journal of Integrative Agriculture. 2018;17(10):2138-2150.
50. Ye X, Li Z, Luo X, Wang W, Li Y, Li R, *et al*. A predatory myxobacterium controls cucumber Fusarium wilt by regulating the soil microbial community. Microbiome. 2020;8(1):1-7.