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Study on the management of *Ralstonia solanacearum* (Smith) with spent mushroom compost

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Abstract
In the present study spent composts of button, oyster mushrooms and their combinations were used both in aqueous and powdered forms for the management of bacterial wilt of tomato caused by *Ralstonia solanacearum*. *In-vitro* and *in-vivo* experiments were conducted. Spent composts significantly reduced bacterial wilt of tomato under screen house conditions and produced larger growth inhibition zones of bacteria on artificial medium. Mixture of oyster and button mushroom restricts bacterial growth up to 8.43 mm followed by button mushroom compost with 7.80 mm. However, mixture of oyster and button composts performed better in the management of bacterial wilt of tomato than applied alone. Likewise, application of powdered spent mushroom composts was more effective than aqueous application. Similarly button mushroom compost was more effective against *R. solanacearum* than oyster compost both in powder and in extracted form. Mixture of oyster and button composts restricts disease severity up to 14.58% when applied as powdered form and up to 27% when applied as aqueous extracts. Moreover, spent mushroom compost improved all agronomic characteristics of tomato plants such as plant height, number of fruits per plant, fresh weight of plant, number of shoots per plant, root length, root weight, and dry plant weight. The ability of spent mushroom compost to reduce disease severity and enhance plant growth could be attributed to the alteration of physical structure of soil and encouragement of plant friendly microbes by the compost.

Keywords: Oyster Mushroom, *Ralstonia solanacearum,* compost, bacterial wilt.

1. Introduction
Tomato *(Lycopersicon esculentum* MILL*),* belonging to family Solanaceae, is an important vegetable crop. It was first cultivated in South America and then widely distributed throughout the world by early explorers [1]. It is a perennial crop, often grown outdoors in temperate climate as an annual crop [2]. Most of the tomato cultivars produce red fruit, but other cultivars with yellow, orange, pink, green, black, purple, or white fruits are also available. Tomato can be used in various ways such as raw in salads, processed as ketchup or tomato soup. Unripe tomatoes can also be breaded and fried, or pickled. Tomato juice is sold as a drink, and is used in cocktails [3]. Similarly, it is an essential part of human diet and is a rich nutritional resource of vitamin C and antioxidants, mainly carotenes, lycopene, phenolics and organic acids [4].
In Pakistan, tomato got a key position amongst other vegetable and is grown throughout the year in different areas. Pakistan and specifically Khyber Pakhtunkhwa (KPK) is favorable for tomato production due to its suitable environmental condition. Currently worldwide tomato production is 100 million tons from an area of 3.7 million ha. The major tomato producing countries are China, United States, Italy, Turkey and India. In Pakistan, the total tomato production is 0.5296 million tons where the area under tomato cultivation is about 52300 ha. Likewise, average tomato production of KPK is 10.5 tones/ha with an area of 12600 ha with a total production 0.2132 tons having an average of 9.8 tones/ha [5].
Inspite of suitable environmental condition, the per hectare tomato yield in KPK is far below from its potential production due to many reasons. Diseases are one of the main factors for low yield. Fungal, bacterial, viral and nematodal diseases cause severe losses to tomato production. The most prominent bacterial diseases include bacterial wilt and canker, soft rot, bacterial speck, southern bacterial wilt, and bacterial spot. Among all these diseases, tomato wilt is of great economic importance. The disease is caused by *Ralstonia solanacearum*, previously known *as Pseudomonas solanacearum.*

R. solanacearum is a Gram-negative strictly aerobic motile, rod. Cells are 0.5-0.7 x 1.5-2.0 µm in size and non-capsulated. The pathogen is catalase positive, oxidase positive, and reduces nitrates. The bacterium does not hydrolyze starch and did not readily degrade gelatin. In broth culture, the organism cannot survive when it contain more than 2% NaCl. For majority of the strains, the optimal growth temperature is 82-90 ^{0}F. Bacterium can be easily isolated on liquid and solid agar growth media. Colonies are white or cream colored, irregularly round fluidal, and opaque [6].

The bacterium has a wide host range and cause infection in more than 200 plant species in 50 families worldwide, including vegetables, ornamentals and fruit plants etc [7]. Annual losses of 15 to 50% both in the lowlands and in the mountain slopes have been recorded.

The disease is characterized as droopiness of younger leaves under favorable condition. In severe cases complete and rapid wilt develops within two to three days after infection. Similarly, plant become desiccated and wilts quickly. In stem vascular system, yellow or light brown sectioning appears. When the disease progress stem becomes dark brown and gradually pith and cortex becomes brown [8]. High temperature (85-95 ^{0}F) favors disease progression. The appearance of sticky, milky exudates from the surface of freshly cut sections is the key diagnostic tool to detect bacterial wilt of tomato [9].

In Khyber Pakhtunkhwa, no detailed research work has been done on the organic management of bacterium wilt. In the current research project, tomato bacterial wilt was managed, using aqueous extracts of different types of spent mushroom compost and their various doses with the objectives to study the effect of spent composts, of oyster and button mushrooms on the management of bacterial wilt of tomato and to evaluate the methods of application (Root dipping and powder mixing in soil) of spent mushroom composts in management of bacterial wilt of tomato.

2. Materials and Methods

2.1 Source of bacterial culture

PCR identified culture of *R. solanacearum* was obtained from the culture bank of the department of plant pathology, The University of Agriculture Peshawar.

2.2 Cultural characteristics

To ensure that the culture obtained from culture bank was pure and un-contaminated its culture characteristics were studied by growing it on LB media (Luria Bertani) for colony margins, color and colony shape etc were observed after the streaked plates were incubated for 48 hours at 27^{0}C.

2.3 Pathogenicity test

To ensure that the stored culture did not lose its pathogenicity, a pathogenicity test was conducted. For this purpose four-week old tomato seedlings were transplanted into porous plastic container. Two days old culture of *R. solanacearum* grown on LB medium was taken and a suspension having 1×10^8 cfu/ml was prepared. Tomato seedlings were inoculated by injecting 3ml bacterial suspension at the crown area of the seedling. Disease symptoms were observed after a few weeks.

2.4 Disease control via spent mushroom compost

Different types and doses of spent mushroom compost were used to test the effect of SMC in the control of tomato bacterial wilt disease. Detailed laboratory and screen house experiments were conducted.

2.5 Preparation of SM compost/ Organic Extract

Spent compost of (button-2012) and (oyster-2013) mushrooms were obtained from Plant Pathology Department mushroom house, The University of Agriculture Peshawar. The compost (oyster 2013, button 2012 and a 1:1 mixture of the two) were first shade dried and then powdered with the help of electric grinder in Animal Nutrition Department, The University of Agriculture Peshawar. The Spent mushroom composts were tested in both powder and aqueous extract form. To make 3-different concentrations of organic extracts, powder of SM composts were soaked in sterile distilled water for 24 hours at the rate of 15g/100ml, 25g/100ml and 35g/100ml [10, 11]. To remove large particles, the organic aqueous extracts were filtered through whatman filter paper.

2.6 *In-vitro* effect of organic extracts on *R. solanacearum*

The experiment was conducted to find out the ability of aqueous extracts of spent mushroom Composts (Button-2012, Oyster-2013, Mixture of Button-2012 and Oyster-2013) to restrict bacterial (*R. solanacearum)* growth. For this purpose paper disc (6 mm) diffusion method was used. Bacterial culture was grown on LB (Luria Bertani) medium and a fresh suspension of (10^8 cfu/ml) was prepared in 0.85% saline. To obtain bacterial culture lawn, LB plates were poured with 100 ul/plate bacterial suspension and evenly spread. Three different organic extract concentrations were prepared as described above and paper discs were soaked in them. Three different concentrations (100ppm, 200ppm, 300ppm) of control antibiotic streptomycin were also prepared and paper discs soaked. Paper discs soaked in water were used as negative control. Paper discs loaded with compost extract, were placed on LB containing plates pre inoculated with bacterial culture. CRD with four replications and two factors was used. The plates were then incubated for 48-hours at 27 ^{0}C and zones of inhibition were recorded. The data obtained were analyzed using statistical package (Statistics 8.1).

2.7 Screen house studies

Screen house experiments were conducted to find out the antimicrobial activity of spent mushroom composts (powder doses and aqueous extracts). In order to inactivate primary inoculum soil was sterilized at 121 ^{0}C and 15 lb for 20 minutes. Locally cultivated tomato cultivar, Rio Grenade, was used for experiment. Fresh bacterial suspension of 1×10^8 cfu/ml in 0.85% saline was prepared. Tomato seedlings were inoculated by root dipping in bacterial suspension for 15 mins [12]. SMC were applied both in powder and extract form. In first experiment, three different concentrations (15g/100ml, 25g/100ml and 35g/100ml) of oyster 2013, button 2012 and mixture of oyster and button (1:1 w/w) were used. Tomato seedlings were dipped in SM Composts extract of all 3-concentrations for 15-mins and then transplanted in pots having two of kg soil, while untreated controls were also included in which sterile distilled water for root dipping. In other experiment in screen house conditions powder of SMC were applied to the pot and amended with soil at the rate of 60g, 120g and 180g/pot before transplantation of seedling. Both positive (streptomycin) and negative (just water added) controls were also kept. CRD with four replicates and two plants per replicate per treatment was used.

2.8 Data collection Parameters

The experiments were ended three months after transplantation and data were recorded on the following parameters.

2.8.1 Disease severity (%)
Data on disease severity were recorded on individual plants of inoculated tomato plants. Disease rating scale of Wright *et al.,* (2005) was used and the values were then averaged for all plants. 0 = no disease symptoms on plants, 1 = disease symptoms less than 50% on plants, 2 = disease symptoms more than 50% on plants and 3 = 100% disease symptoms or plant totally dead [13]. Disease severity scale values were converted to % disease severity using the following formula [14].

$S = 100\sum n/N \times 3$

S = % disease severity

$\sum n$ = individual ratings summation

N = total no of plants assessed

3 = highest score on disease severity scale

2.8.2 Plant height (cm)
Tomato plants were uprooted after 2 months of their transplantation and their height were then measured individually (cm) with the help of measuring tape.

2.8.3 Number of fruits (tomato) per plant
Tomato fruits were harvested three times during the whole season and data were noted for each plant in each treatment.

2.8.4 Fresh plant weight (g)
Fresh weight of each uprooted plant was recorded with the help of digital balance.

2.9 Data analysis
Data were analyzed by using statistical package (Statistix 8.1). Different parameters were treated as dependent variables, while treatments were considered as independent variable. Means were separated by least significant difference (LSD) test. [15].

3. Results
3.1 Characterization of *Ralstonia solanacearum*
R. solanacearum colonies grown on Luria Bertani (LB) culture medium were white or cream colored, fluidal, irregularly round and opaque. Wilting of four-week old tomato seedlings confirmed the pathogenicity of the bacteria.

3.2 *In-vitro* antibacterial activity of SM compost against *R. solanacearum*
Significant ($P \leq 0.05$) differences between spent mushroom composts and their concentrations were noticed when tested against *R. solanacearum* under *in-vitro* conditions. Maximum zone of inhibition (minimum bacterial growth) recorded for mixture of oyster and button (COB) was 8.43 mm followed by button 7.80 mm and oyster producing 7.30 mm inhibition zones. Zones of inhibition produced by spent mushroom composts were compared with water negative control and streptomycin sulphate antibiotic were used as standard. Aqueous extract concentrations of all spent mushroom composts showed significant differences ($P \leq 0.05$) in zones of inhibition on LB culture medium. Maximum inhibition zone of 7.18 mm was noticed by extract three (E_3= 35g/100ml) followed by extract two (E_2 = 25g/100ml) with inhibition zone of 6.85 mm. significant differences ($P \leq 0.05$) were observed between interactions of spent mushroom compost and their various concentrations. Maximum zone of inhibition (8.87 mm) was recorded when mixture of oyster and button were used at a concentration of 35g/100ml followed by 8.47 mm when used at a concentration of 25g/100ml of water. Streptomycin sulphate used as standard recorded 11.2 mm inhibition zone when used at the rate of 300 ppm. Water control recorded 0 mm zone of inhibition.

Table 1: Effect of aqueous concentration of spent mushroom compost on growth (mm) of *Ralstonia solanacearum* on LB Medium

Extract	COMPOST					Mean
	Oyster (C_1)	Button (C_2)	COB (C_3)	Streptomycin (ppm)	Water Control	
E_1(15 g/ 100 ml)	6.95 i	7.40 h	7.95 fg	10.12 c	0.0 j	6.48 c
E_2 (25 g/ 100 ml)	7.27 i	7.82 fg	8.47 e	10.7 b	0.0 j	6.84 b
E_3 (35 g/100 ml)	7.65gh	8.17 ef	8.87 d	11.22 a	0.0 j	7.18 a
Mean	7.30 d	7.80 c	8.43 b	10.68 a	0.0 e	

LSD value for compost 0.22

LSD value for concentration 0.17

LSD value for compost x concentration 0.39

3.3 *In-Vivo* studies (Screen House)
3.3.1 Effect of mushroom compost on number of plant shoots
Significant differences ($P \leq 0.05$) were found in number of shoots when three different powder doses of oyster and button mushroom composts and their mixture were applied in powder form. Maximum numbers of shoots (18.0) were observed when mixture of oyster and button mushroom compost (COB) was applied, followed by button with 15.6 numbers of shoots as shown in Table 2. Powder dose three (P_3 = 180g/pot) was found best with maximum number of shoots 17.35 followed by powder dose two (P_2 = 120g/pot) with 15.8 number of shoots per plant. Interaction between spent mushroom compost and their powder doses was also found to be significant ($P \leq 0.05$). C_3 P_3 (P_3 = 180g/pot) produced maximum number of shoots (19.75) followed by C_3 P_2 (120g/pot) with 17.75 number of shoots per plant. On the other hand, streptomycin resulted in maximum number of plant shoots (23.25) when applied with a concentration of 300 ppm, while water control produced minimum number of shoots (9.75) per plant.

The effect of Spent mushroom compost on number of plant shoots were obvious ($P \leq 0.05$) when applied at various concentrations in aqueous extract form. Maximum number of shoots (9.4) per plant were observed in pots, treated with mixture of button and oyster extract (COB) followed by button compost extract (8.0) as shown in Table 3. Different concentrations of compost extract increased number of shoots significantly. E_3 (35g/100ml) revealed maximum number of shoots (10.25) among the tested concentrations followed by E_2 (25g/100ml) with 8.45 number of shoots per plant. In case of interaction combinations, no significant differences ($P \leq 0.05$) were observed C_3E_3 (COB @ 35g/100ml) resulted in highest number of shoots (11.25) followed by C_2 E_3 (button @ 25g/100ml) with 9.75 shoots per plant. Maximum numbers of shoots per plant (16.75) were observed when streptomycin was applied at the rate of 300 ppm concentration. While minimum numbers of shoots per plant (4.25) were observed on negative control (water).

Table 2: Effect of powder application of spent mushroom compost on number of shoots of tomato plant inoculated with *Ralstonia solanacearum*

Powder	Compost					Mean
	Oyster (C_1)	Button (C_2)	COB (C_3)	Water Control	Streptomycin (ppm)	
P_1 (60g)	12.5 h	14.5 g	16.75 def	9.75 i	20.25 bc	14.75 c
P_2 (120g)	14.25 g	15.25 fg	17.75 d	10.25 i	21.5 b	15.80 b
P_3 (180g)	15.75 efg	17.25 de	19.75 c	10.75 i	23.25 a	17.35 a
Mean	14.1 D	15.66 c	18.0 b	10.25 e	21.6 a	

LSD value for compost 0.88
LSD value for concentration 0.68
LSD value for compost x concentration 1.53

Table 3: Effect of aqueous concentration of spent mushroom compost on number of shoots of tomato plant inoculated with *Ralstonia solanacearum*

Extract	Compost					Mean
	Oyster (C_1)	Button (C_2)	COB (C_3)	Water Control	Streptomycin (ppm)	
E_1 (15 g/ 100 ml)	5.75 ghi	6.5 fgh	7.75 f	4.25 i	12.25 c	7.30 c
E_2 (25 g/ 100 ml)	6.5 fgh	7.75 efg	9.25 de	4.25 i	14.5 b	8.45 b
E_3 (35 g/100 ml)	8.25 ef	9.75 de	11.25 cd	5.25 hi	16.75 a	10.25 a
Mean	6.8 d	8.0 c	9.4 b	4.6 e	14.5 a	

LSD value for compost 1.18
LSD value for concentration 0.92
LSD value for compost x concentration 2.05

3.3.2 Effect of mushroom compost on root length (cm) of tomato

Root length increased significantly ($P \leq 0.05$) when three different powder doses of oyster and button mushroom composts and their mixture were applied in powder form. Maximum root length (14.37 cm) was recorded by Mixture of oyster and button spent mushroom compost (COB) followed by button with root length of 12.43 cm. Powder dose three (P_3 = 180g/pot) was found effective with maximum root length of 14.35 cm followed by powder dose two (P_2 = 120g/pot) with root length of 13.20 cm. Interaction between spent mushroom compost and their powder doses was also found to enhance root length significantly ($P \leq 0.05$). Maximum root length (16.40 cm) was observed in C3 P_3 (P_3 = 180g/pot) followed by C_3 P_2 (120g/pot) with root length of 14.32 cm. Streptomycin resulted in maximum root length (20.35 cm) when applied with a concentration of 300 ppm, while water control showed minimum root length (8.85 cm) as shown in Table 4.

Effect of mushroom compost aqueous extracts on root length was also obvious at five percent level of probability. Maximum root length (11.0 cm) was recorded in pots, treated with mixture of button and oyster extract (COB) followed by button compost extract (9.1 cm) as shown in Table 5. Significant differences were observed when various concentrations spent mushroom composts were used. E_3 (35g/100ml) showed maximum root length (11.02 cm) followed by E_2 (25g/100ml) with 9.85 cm root length. In case of composts*concentrations interaction combinations C_3E_3 (COB @ 35g/100ml) resulted in maximum root length (13.03 cm) followed by C_3 E_2 (COB @ 25g/100ml) with a root length of 10.93 cm. Standard antibiotic streptomycin, enhanced root length up to 17.09 cm when applied @ 300 ppm concentration. While negative control (water) had minimum root length of 5.4 cm as shown in Table 2(b).

Table 4: Effect of powder application of spent mushroom compost on root length (cm) of tomato plant inoculated with *Ralstonia solanacearum*

Powder	Compost					Mean
	Oyster (C_1)	Button (C_2)	COB (C_3)	Water Control	Streptomycin (ppm)	
P_1 (60g)	10.07 hi	11.1 g	12.40 f	8.90 j	18.32 c	12.16 c
P_2 (120g)	10.92 gh	12.32 f	14.32 E	9.12 ij	19.32 b	13.20 b
P_3 (180g)	12.27 f	13.90 e	16.40 d	8.85 j	20.35 a	14.35 a
Mean	11.06 D	12.43 c	14.37	8.96 e	19.33 a	

LSD value for compost 0.57
LSD value for concentration 0.44
LSD value for compost x concentration 0.99

Table 5: Effect of aqueous concentration of spent mushroom compost on root length (cm) of tomato plant inoculated with *Ralstonia solanacearum*

Extract	Compost					Mean
	Oyster (C_1)	Button (C_2)	COB (C_3)	Water Control	Streptomycin (ppm)	
E_1 (15 g/ 100 ml)	6.68 hi	7.73 g	9.04 f	5.5 j	14.93 c	8.79 c
E_2 (25 g/ 100 ml)	7.57 gh	8.92 f	10.93 e	5.75 ij	16.09 b	9.85 b
E_3 (35 g/100 ml)	8.88 f	10.64 e	13.03 d	5.4 j	17.09 a	11.02 a
Mean	7.71 d	9.1 c	11.0 b	5.6 e	16.04 a	

LSD value for compost = 0.56
LSD value for concentration = 0.43
LSD value for compost x concentration. =0.97

3.3.3 Effect of mushroom compost on root weight (gm) of tomato

All the tested mushroom composts significantly ($P \leq 0.05$) enhanced the fresh root weight of tomato plants. Mixture of oyster and button spent mushroom compost showed maximum root weight of 24.1 grams, followed by button with root weight of 21 grams. Powder dose three (P_3 = 180g/pot) had maximum root weight of 24.22 grams followed by powder dose two (P_2 = 120g/pot) with root weight of 21.11 grams. In case of interaction combinations, mushroom composts and their powder doses significantly ($P \leq 0.05$) increased root weight of tomato plant. Maximum root weight (28.25 grams) was observed in C3 P_3 (P_3 = 180g/pot) followed by C_2 P_3 (180g/pot) with root weight of 25.1 grams. Positive control streptomycin enhanced root weight up to 33.57 grams, when applied with a concentration of 300 ppm, while negative control (water) showed minimum root weight (10.5 grams) as shown in Table 6.

All the tested levels of spent mushroom compost aqueous extracts significantly ($P \leq 0.05$) increased the root weight Effect of mushroom compost aqueous extracts on root length was also obvious at five percent level of probability. Maximum root weight 24.8 grams was recorded in pots, treated with mixture of button and oyster extract (COB) followed by button compost extract (20.08 grams). Significant differences were observed when various concentrations of spent mushroom composts were used. E_3 (35g/100ml) showed maximum root weight of 23.1 grams followed by E_2 (25g/100ml) with 19.34 grams root weight. Interaction combinations showed non-significant increase in root weight. C_3E_3 (COB @ 35g/100ml) resulted in maximum root weight (29.75 grams) followed by C_3E_2 (COB @ 25g/100ml) with a root weight of 25.75 grams. Positive control streptomycin, increased root weight up to 27 grams when applied @ 300 ppm concentration. While negative control (water) had minimum root weight of 11 grams as shown in Table 7.

Table 6: Effect of powder application of spent mushroom compost on root weight of tomato plant inoculated with *Ralstonia solanacearum*

Powder	Compost					Mean
	Oyster (C_1)	Button (C_2)	COB (C_3)	Water Control	Streptomycin (ppm)	
P_1 (60g)	15.32 i	16.65 hi	20.25 g	10.5 j	26.37 cd	17.82 c
P_2 (120g)	18.8 gh	21.22 fg	23.82 def	11 j	30.72 b	21.11 b
P_3 (180g)	23.17 ef	25.1 de	28.25 bc	11 j	33.57 a	24.22 a
Mean	19.1 d	21.0 c	24.1 b	10.8 e	30.2 a	

LSD value for compost 1.52
LSD value for concentration 1.18
LSD value for compost x concentration 2.64

Table 7: Effect of aqueous concentration of spent mushroom compost on root weight of the plants inoculated with *Ralstonia solanacearum*

Extract	Compost					Mean
	Oyster (C_1)	Button (C_2)	COB (C_3)	Water Control	Streptomycin (ppm)	
E_1 (15 g/ 100 ml)	10.75 i	14.75 fg	18.75 ef	11 g	19.25 defg	14.90 c
E_2 (25 g/ 100 ml)	15.75 fgh	20.75 cde	25.75 abc	11.72 g	22.75 bcde	19.34 b
E_3 = 35 g/100 ml	20.75 cde	24.75 abc	29.75 a	13.25 g	27 ab	23.1 a
Mean	15.75 c	20.1 b	24.75 a	12.0 d	23.0 ab	

LSD value for compost 3.31
LSD value for concentration 2.56
LSD value for compost x concentration 5.74

3.3.4 Effect of mushroom composts on dry weight (gm) of tomato plants

Significant effect of spent mushroom composts on dry weight of tomato plants were noticed at five percent level of probability. Mixture of oyster and button mushroom composts resulted in highest dry plant weight (89.7 grams) followed by button compost with dry plant weight of 86.00 grams as shown in Table 8. Dry Plant weight differed significantly ($P \leq 0.05$) when three different powder doses of oyster and button mushroom composts and their mixture were applied in powder form. Powder dose three (P_3 = 180g/pot) enhanced dry plant weight (88.65 grams) followed by powder dose two (P_2 = 120g/pot) with 84.60 grams dry plant weight. Interaction between spent mushroom compost and their powder doses was also found to be significant ($P \leq 0.05$). C_3 P_3 (P_3 = 180g/pot) resulted in dry plant weight up to 97.75 grams followed by C_2 P_3 (180g/pot) with weight of 96.75 grams. Positive control streptomycin resulted in maximum dry plant weight (105.5 grams) when applied at a concentration of 300 ppm, while water control showed minimum dry plant weight of 42.75 grams.

All mushroom compost aqueous extracts significantly ($P \leq 0.05$) increased dry weight of tomato when used alone as well as in combined form in different concentrations. Maximum dry plant weight (78.0 grams) was recorded in pots, treated with mixture of button and oyster extract (COB) followed by button compost extract (60.6) as shown in Table 9. Significant differences were observed when various concentrations of spent mushroom composts were used. E_3 (35g/100ml) showed maximum dry plant weight (70.0 grams) followed by E_2 (25g/100ml) with 63.65 grams. In case of interaction combinations, C_3E_3 (COB @ 35g/100ml) resulted in maximum dry plant weight (81.25 grams) followed by C_3E_2 (COB @ 25g/100ml) with 77.75 grams dry plant weight. Standard antibiotic streptomycin enhanced dry plant weight up to 94.75 grams when applied @ 300 ppm concentration. While negative control (water) had minimum dry plant weight of 36.51 grams as shown in Table 9.

Table 8: Effect of powder application of spent mushroom compost on dry weight (gm) of tomato plant inoculated with *Ralstonia solanacearum*

Powder	Compost					Mean
	Oyster (C_1)	Button (C_2)	COB (C_3)	Water Control	Streptomycin (ppm)	
P_1 (60g)	72.75 e	73.5 e	80.75 d	42.75 g	87.25 c	71.40 c
P_2 (120g)	82.75 d	87.75 c	90.75 c	53 f	98.75 b	84.20 b
P_3 (180g)	87.75 c	96.75 b	97.75 b	55.5 f	105.5 a	88.65 a
Mean	81.0 d	86.0 c	89.7 b	50.4 e	97.1 a	

LSD value for compost 2.23
LSD value for concentration 1.73
LSD value for compost x concentration 3.87

Table 9: Effect of aqueous concentration of spent mushroom compost on dry weight (gm) of tomato plant inoculated with *Ralstonia solanacearum*

Extract	Compost					Mean
	Oyster (C_1)	Button (C_2)	COB (C_3)	Water Control	Streptomycin (ppm)	
E_1 (15 g/ 100 ml)	42.75 g	55.75 f	75.25 cd	37.25 h	72 d	56.60 c
E_2 (25 g/ 100 ml)	61.25 e	60.5 ef	77.75 bc	40.25 gh	78.5 bc	63.65 b
E_3 (35 g/100 ml)	71.5 d	65.75 e	81.25 b	36.5 h	94.75 a	70.0 a
Mean	58.5 c	60.6 c	78.0 b	38.0 d	81.7 a	

LSD value for compost 3.06
LSD value for concentration 2.37
LSD value for compost x concentration 5.30

4. Discussion

The present study was conducted to test the effectiveness of oyster and button spent mushroom compost for the management of bacterial wilt of tomato caused by *Ralstonia Solanacearum.* SM compost are the organic remains (organic material) after mushroom harvest and is known for its ability to improve chemical, physical, and biological properties of soil.SM compost activate soil microbially and suppress root and soil borne pathogen [16]. In the current research work three different types of SM composts (Oyster 2013, Button 2012 and 1:1 mixture of both) were used both in aqueous extract form (3-different concentrations) and powder form (3-different doses).

Findings of the current research work revealed the effectiveness of spent mushroom compost in extracted form against bacterial growth under laboratory conditions. Extract of mixture (button and oyster at 1:1 ratio) spent mushroom compost resulted in larger zone of inhibition (8.4 mm) followed by the SM compost of button under laboratory conditions. Our results are in line with those of Mumtaz *et al*. *Erwinia carotovora subsp carotovora*, the causal agent of bacterial soft rot in tomato, produced reduced growth on nutrient medium when treated with paper disc dipped in the extract of SMC of oyster and button [17]. Similarly Farhad also found spent mushroom compost effective in reducing bacterial growth in extracted form [18].

An inverse relationship was found between compost rate and bacterial growth i.e. the greater the dose of spent mushroom compost, lower was bacterial growth. Mixture of oyster and button compost in 1:1 ratio significantly decreased *Ralstonia solanacearum* multiplication. Mumtaz *et al.* found mixture of oyster and button spent mushroom to be more effective than either alone, in reducing plant mortality, high leaf index and plant height of rough lemon (*Citrus jambhiri* L*)* [17]. The chemical analysis of the mixture showed higher N, P and K values. Effectiveness of button spent mushroom compost over oyster spent mushroom may be possibly due to variable physical, chemical and biological nature of both the SMC. Analysis of spent mushroom compost revealed the presence of antagonistic microbial population such as Pseudomonas and Bacillus. *Pseudomonas aeruginosa* which was found in spent compost of mushroom reduced the multiplication of *Pyricularia grisea* [19]. Similarly, reduced conidial germination (98% reduction in growth) of *Venturia inaequalis* was noticed when treated with water extract of spent mushroom compost. Spore germination of artillery fungi was restricted with the help of spent mushroom compost [20]. Spent compost of mushroom enhances natural defense system of plant in extracted form by triggering resistance gene [21].

The result of the screen house study indicated that spent mushroom compost when used in combination (button and oyster 1:1) was better than either button or oyster compost alone. Moreover button spent compost was more effective both in extracted and powdered form than oyster spent mushroom compost. Increasing level of SM compost reduced disease severity i.e. powder dose three, i.e. 180g per pot and aqueous extract dose three of 35gm/100ml were more effective as compared to dose one and two. Button SM compost was more effective than oyster in disease suppresiveness both in powder dose and aqueous extract form, possibly due to its variable nature. Application of SM compost at higher rates both in aqueous extract and powder form (35gml100ml and180gm/pot) were more effective in disease suppression and improved agronomic characteristics may be due to the presence of higher levels of antagonistic microbes and active compounds. Our findings were in line with those of Israr (2012) in terms of reducing disease severity % under screen house conditions [22]. Lamondia *et al* noted a decrease in early dying disease of potato by growing it in soil amended with spent compost mushroom. Incidence and growth of *Botrytis cinerea* and conidial germination of *Cochliobolus carbonum* was effectively controlled by the application of SMC [23, 24]. SM compost amended soils have large population of antagonistic microbes like *Bacillus subtilis, B. licheniformis, Pseudomonas aeruginosa, S. albidoflavus* which contributes n minimizing disease severity.

In addition to reducing bacterial growth on nutrient medium and decreasing in disease severity under laboratory and screen house conditions respectively SM compost was also found effective in improving agronomic characteristics like fresh plant weight, root length, plant height, dry plant weight, number of fruits and shoots per plant of tomato plant. Those plants which received mixture of button and oyster (in extracted and powdered form) spent mushroom exhibited

improved growth pattern.SM compost in powder form increases the water holding capacity of growth medium thus resulting in better seed germination due to reduction in desiccation of the medium. Romaine and Holcomb also reported better performance of compost in powder form as compared to coarse compost in tomato growth response evaluation [25]. SM compost enhance crop yield by increasing nutrient uptake, soil porosity, water holding capacity, antagonistic microbial population. Spent mushroom compost improve soil structure, water holding capacity, soil aeration, enrich soil with nutrient (NO_3, N, Mg, K, P and Ca etc) and microbial activity like (*Bacillus licheniformis, B. subtilis, Pseudomonas aeribacillus, P. aeruginosa and Streptomyces albidoflavus).* Kadiri and Mustapha observed significant increase in fresh plant weight, dry plant weight, leaf area, number of leaves per plant, number of fruit/pod/seed per plant and stem growth in tomato and cowpea growth in spent mushroom compost amended soil [26].

5. Conclusion and Recommendations

Different spent mushroom like button and oyster their 1:1 mixture, used in this study was found most effective in reducing bacterial growth and disease severity. Mixture of button and oyster spent mushroom resulted in better performance in term of reducing disease severity by inhibiting bacterial growth. Efficacies of spent mushroom compost were increased with increasing level of spent mushroom compost in powdered and extracted form. Spent mushroom substrate was more effective in powder form as compared to use in extracted form. Spent mushroom compost should be integrated as component to integrated disease management strategies. Spent mushroom substrate can be used both in fine powdered form and as water extract. Further studies are required on the biochemical assay of spent mushroom compost. Studies are also required to find its effect on the management of other plant diseases.

6. References

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