

Role of Combination among Bioagents, Compost and Soil Solarization on the Management of Some Root Infecting Fungi Responsible for Crown and Root-Rot Diseases of Strawberry

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ABSTRACT

The antagonistic bacteria, i.e. *B. megaterium*, *B. pumilus*, *B. subtilis*, *Pseudomonas fluorescens* and *P. putida* isolated from the rhizospheric soil of strawberry plants were screened for their efficacy against the pathogenic fungi, i.e. *Fusarium solani*, *Macrophomina phaseolina* and *Rhizoctonia solani*, responsible for causing crown and root-rot diseases of strawberry, *in vitro* and *in vivo*. In general, *P. fluorescens* followed by *megaterium* were the most efficient ones in reducing the linear growth of the tested three pathogenic fungi than the other isolates.

Sterilized filtrate of the tested compost (compost tea) resulted in significant reduction to the linear growth of the tested three fungi compared with the control treatment. This reduction was gradually increased by increasing its concentration.

The Average of the soil temperature at depths of 15 and 25 cm. was 59.7 and 51.3 °C during the period of soil solarization. The pathogenic three fungi failed to re-isolate from the infected segments of solarized roots for 40 days at 15 and 25 cm. depths. In addition, low frequency was recorded in case solarized crown segments, especially at 25 depths.

Plot experiment revealed that the tested bioagents, i.e. *B. megaterium* and *P. putida*, compost and soil solarization, each alone or in different combination, caused significant reduction to the infection by the mixed inoculums of the three causal fungi (dead plants and root-rot severity) with significant increase to the produced fruit yield and their total soluble solids (TSS) compared with the control treatment.

Keywords: Strawberry, bacterial bioagents, compost, crown and root-rot, fruit yield, management, soil solarization, total soluble solids (TSS).

INTRODUCTION

Strawberry (*Fragaria × ananassa*) is one of the most important and delicious untraditional crops in Egypt for local consumption and exportation. It is of high cash return from foreign exchange. It is liable to infection by many bacterial, viral diseases in addition to nematode infection and physiological disorder. Strawberry is grown in most arable regions of the world. The crop is enjoyed by millions of people in all kinds of climates including temperate, Mediterranean, subtropical and taiga zones (Mass, 1998). However, a fungal soil borne fungus pose serious threat to commercial strawberry production worldwide and causes severe economic losses (Attia et al., 1989; Mostafa et al., 1999; Fang et al., 2011 and 2012; Juber et al., 2014 and Abada et al., 2014 and Abada and Hassan, 2017). During the last decade, many

complaints have been received from strawberry growers due to the death of strawberry plants due to the infection by soil borne fungi just after transplanting until end of the growing season.

The control of soil borne diseases is currently accomplished primarily through the use of soil fumigation by methyl bromide as well as fungicides and, to somewhat, resistant cvs. However the frequent and discriminate use of soil fumigation and fungicides leads to atmosphere pollution and create imbalance in the microbial community, which may be unfavorable to the activity of beneficial organisms and may lead to development of resistance strains of the pathogen (Martin and Bull, 2002). In recent years biological control, soil solarization and compost has become a promising safer and ecologically acceptable alternative to chemical control in the management

of soil-borne diseases (Fahim et al., 1990 ; Santoyo et al., 2012 ; Abada and Ahmed, 2014 ; Abada et al., 2014 and Abada and Hassan, 2017). Among the bacterial bioagents, genera *Bacillus* and *Pseudomonas* received more attention than many other bacterial groups (Santoyo et al., 2012; Abada et al., 2014 and Abada and Hassan, 2017).

Most strawberry fruits are consumed mainly as fresh or canned; therefore disease management rather than chemical control must be used. In recent years, biological control has emerged as an alternative and most promising means of the management of plant pathogens.

Biocontrol of the causal of strawberry soil-borne diseases could be achieved by either promoting the native antagonists such as that found in compost to reach a density sufficient to suppress pathogen(s) or by introducing alien antagonists. Among the several antagonists tested by various scientists, genera of *Bacillus* and *Pseudomonas* have been found effective in inhibiting the causal of strawberry crown, root-rot and wilt (Abada et al., 2014; Barakat et al., 2014; Ragab et al., 2015 and Abada and Hassan, 2017). Therefore, introduce of several antagonists against these pathogens seems to hold great promise to suppress the disease and have been found effective in inhibiting the growth of the tested fungi under in vitro conditions.

The present investigation aimed to investigate the role of the combination among bacterial bioagents, compost and soil solarization in management of crown and root-rot diseases of strawberry.

MATERIALS AND METHODS

Source of Fungi Responsible for Crown and Root-Rot Diseases of Strawberry as Well as the Bacterial Bioagents

Pathogenic isolates of *Fusarium solani*, *Marophomina phaseolina* and *F. solani* as well as the bacterial bioagent, i.e. *B. megaterium*, *B. pumilus*, *B. subtilis*, *Pseudomonas fluorescens* and *P. putida* were kindly provided by Dr. K.A. Abada, Prof. of Plant Pathol., Fac. Agric., Cairo Univ.

Effect of the Culture Filtrate of the Tested Bioagents on the Linear Growth of the Tested Fungi

The effect of the culture filtrate of *B. megaterium*, *B. pumilus*, *B. subtilis*, *P. fluorescens* and *P. putida* on the linear growth of three pathogenic fungi was

studied as a method given by Dennis and Webster (1971).

Nutrient medium (350 ml.) were put in each 500 ml flask and sterilized by steamer for three successive days. The medium was inoculated with a loop of any of the tested five bacterial bioagents taken from two days old culture. Inoculated flasks were incubated at $30 \pm 1^\circ\text{C}$ on a rotary shaker at 200 rpm for two days. The culture filtrate was filtered through Whitman No.1 filter paper and collected in a flask. The culture filtrate of each bioagent was sterilized using a $0.25\mu\text{m}$ syringe filter. The sterilized filtrate was mixed with sterilized PDA medium just before solidification in different proportion (10, 20, 40 and 60%). PDA medium plus culture filtrate of the bioagent(s) were poured into sterilized Petri-dishes (20 ml/plate).

After solidification the Petri-dishes were carefully inoculated with 5 mm. discs of any of the test pathogens cut from five days old cultures. PDA plates inoculated with the tested pathogens, but not amended with culture filtrate were maintained as control. Dishes were then incubated in an incubator at $30 \pm 1^\circ\text{C}$. Five plates were used for each treatment. Observation was done daily on the fungal growth and the linear growth of the tested fungi was recorded. Inhibition percentage of the mycelial growth of tested pathogens was calculated by the formula:

$$I \% = (C - T) / C \times 100$$

Where;

I = the inhibition in the growth of the test pathogen,

C = Linear growth of the pathogen (mm) in control and

T = Linear growth of the pathogen (mm) in the treatment.

Effect of Filtrate of Compost Tea on the Linear Growth of the Causal Pathogens

Plant compost (2.0 kg.) was soaked overnight in five liter water in a plastic container then filtrates through two layers of cheesecloth then by Whitman 1 filter paper and collected in a flask. Compost tea filtrate was sterilized using a $0.25\mu\text{m}$ syringe filter. The sterilized filtrate was mixed with sterilized PDA medium just before solidification in different proportion, i.e. 10, 20, 40 and 60%). The medium plus compost tea filtrate was poured into sterilized Petri-dishes (20 ml/plate). After solidification the

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Petri-dishes were carefully inoculated with 5 mm. discs of any of the test pathogen cut from five day old culture. PDA plates inoculated with the test pathogen, but not amended with compost tea filtrate (normal PDA) were maintained as control. Plates were then incubated in an incubator at $28 \pm 2^\circ\text{C}$. Five replications were prepared for each treatment. Observation was done on the linear growth of the tested fungi, measured and the average was recorded. Inhibition percentage of the mycelial growth of test pathogens was calculated as mentioned before.

Preparation the Inoculums of the three Pathogenic Fungi and the Two Bioagents

The tested three pathogenic fungi were grown on sterilized corn-sand medium in 500 ml. glass bottles for two weeks at $28 \pm 2^\circ\text{C}$. Also, the two antagonistic bacterial bioagents were grown in 300 ml meat broth (10 g meat extract, 10 g peptone, 5 g tryptone, and 5 g glucose in 1 liter water) for two days at $28 \pm 2^\circ\text{C}$ and the concentration of 1×10^6 cfu / ml. water was prepared from each bioagent.

Effect of Soil Solarization on the Viability of the Three Pathogenic Fungi

Plastic pots contained formalin disinfected soil were infested with the inoculum of the tested pathogenic fungi, i.e. *F. solani*, *M. phaseolina* and *R. solani*, each alone, irrigated and left for five days before transplanting strawberry transplants. Five Frigo transplants (Festival CV.) Were transplanted in each pot and ten pots were used for each fungus. The grown plants were pull-off, three months after transplanting and the infected crowns and main roots were cut into small parts (0.5 cm. long) and five segments from the crowns and five segments from the roots were put in nylon socks and closed well. Nylon socks containing the infected crowns and roots, each alone were buried in the soil of the plots (contained soil of about 55 % water holding capacity) at 15 and 25 cm. depths. Five nylon socks were put in each depth in each plot. Three plots were used for each fungus. The plots were covered with plastic sheets (50 μ thick) for 40 days beginning from end of June, 2018. Also, five nylon socks containing the infected crowns and roots, each alone were buried at the same two depths put without covering with plastic sheets as control treatment (un-mulched soil). A soil thermometer was put at each depth of three plots of each treatment and the temperature was taken daily at 2.0 pm and the average degree of

temperature for each depth was calculated and recorded. After the elapse time, nylon socks collected and taken for re-isolation from both solarized crowns and roots and control treatment. The crowns and the roots of each fungus treatment were sterilized in 2% sodium for two minutes. The tissue pieces were subsequently washed in three changes of sterile water to eliminate excess sodium chlorite and then the five pieces from any of the crowns and roots were transferred onto PDA medium in Petri-dishes for each fungus. Five Plates for each of crowns and roots of each fungus were incubated at $28 \pm 2^\circ\text{C}$ and observed periodically for growth of the fungi. The emerged fungi from the infected crowns and roots were counted and the average was recorded for each treatment.

Effect of the Combination among the Bacterial Bioagents, Compost and Soil Solarization on Management of Crown and Root-Rot Diseases

In the present investigation, transplants of Festival strawberry cv. were brought from Strawberry Development Center, Fac. of Agric., Ain Sahms Univ., where previously grown in nursery fumigated with methyl-bromide. The antagonistic *B. megaterium* and *P. fluorescens* were tested for their efficiency in management of crown and root-rot diseases in combination with compost tea and soil solarization. The experiment was carried out in plots in the open air in experimental stat. of Plant. Pathol. Dept., Fac. Agric., Cairo Univ. Plots (one m²) contained moist Nile silt soil. The soil was disinfected by 5% formalin solution (Abada, 1986). The upper 20 cm, layer of each plot (1m²), was infested with the mixture of the inoculums of the three pathogenic fungi at the rate of 2 % inoculum level for each fungus. The plots received the following treatments:

- Infested plots with the mixture of the three pathogenic fungi (2% inoculums level from each pathogen) received 2 Kg. of compost.
- Infested plots with the mixture of the three pathogenic fungi were infested with the bioagent *B. megaterium* at the rate of 5 l. / plot (1×10^6 cfu / ml. water).
- Infested plots with the mixture of the three pathogenic fungi were infested with the bioagent *P. fluorescens* at the rate of 5 L. / plot (1×10^6 cfu/ ml. water).
- Infested plots with the mixture of the three pathogenic fungi were solarized by plastic

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sheet (50 μ thick) during first of August, 2018 for 40 days according to Abada and Hassan (2017).

- Two kg. Compost were added to the infested plots with the mixture of the three pathogenic fungi and infested with the bioagent *B. megaterium* at the rate of 5 L. / plot (1x06 cfu/ ml. water).
- Two kg. Compost were added to the infested plots with the mixture of the three pathogenic fungi and infested with the bioagent *P. fluorescens* at the rate of 5 L. / plot (1x06 cfu/ ml. water).
- Two kg. Compost were added to the infested plots with the mixture of three pathogenic fungi and solarized by plastic sheet (50 μ thick) during first of August, 2018 for 40 days.
- Infested plots with the mixture of the three pathogenic fungi were infested with the bioagents *B. megaterium* and *fluorescens* at the rate of 2.5 L. / plot (1x06 cfu/ ml. water) from each bioagent.
- Solarized infested plots with the three pathogenic fungi were infested with the bioagent *B. megaterium* at the rate of 5 L. / plot (1x106 cfu/ ml. water).
- Solarized infested plots with the three pathogenic fungi were infested with the bioagent *P. fluorescens* at the rate of 5 L. / plot (1x06 cfu/ ml. water).
- Solarized infested plots with the three pathogenic fungi received two kg. Compost and infested with both bioagents *B. megaterium* and *P. fluorescens* at the rate of 2.5 L. / plot (1x06 cfu/ ml. water) from each bioagent.
- Infested plots with the mixture of the three pathogenic fungi were kept without any additional treatments (Infested control).
- Un-infested plots with any pathogen were kept without any additional treatments (un-infested control).

After the previous treatments the plots were irrigated and left for one week then transplanted with Festival strawberry cv. at the rate of 16 transplants/ plot at October 20, 2018. Three plots were used for each treatment. The plots were irrigated when it was necessary and fertilized with recommended doses as recommended by Min. of Agric. and Land reclamation. The produced fruits were harvested periodically and the final fruit yield (g./ plant)

was calculated and recorded. Also, total soluble solids (T.S.S.) of five randomly fruits were measured using a hand refractometer (Abada and Hassan, 2017). In addition, visual assessment was carried out for the vigor growth of the grown strawberry plants, where: += Poor growth, ++= Moderate growth, +++= Very good growth and ++++ = Excellent growth.

DISEASE ASSESSMENT

The number of dead plants during the experiments was recorded for each treatment. Also, a visual assessment for crown and root-rot was assessed based on a 0–4 disease severity scale (Phillips and Golzar (2008), where: 0 = no crown and root tissue discoloured; 1 = <25% crown and root tissue discoloured; 2 = \geq 25, <50% crown and root tissue discoloured; 3 = \geq 50, <75% crown and root tissue discoloured; 4 = \geq 75% crown and root tissue discoloured. Disease severity was assessed on four randomly plants in each plot, five months after transplanting and the average was recorded as follows:

$$\text{Disease severity \%} = \frac{\sum (nxv)}{3 N} \times 100$$

Where: n = Number of infected crowns and roots in each category.

v = Numerical values of each category.

N = Total number of the infected crowns and roots.

STATISTICAL ANALYSIS

The obtained data were statistically analyzed using the standard procedures for complete randomized block and split designs as reported by Snedecor and Cochran (1967). The obtained averages were compared at the level of 5% level using least significant differences (L.S.D.) according to Fisher (1948).

RESULTS

Effect of the Tested Bioagents on the Linear Growth of the Tested Pathogens

Results presented in Tables (1, 2 and 3) indicate that the five tested bacterial bioagents resulted in significant reduction to the linear growth of the pathogenic fungi, i.e. *F.solani*, *M.phaseolina* and *R.solani*, 4 days after incubation at 28 \pm 2 $^{\circ}$ C compared with control treatment. This inhibitory was gradually increased by increasing the incorporated concentration. In addition, the three pathogenic fungi were greatly affected by

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both *P. fluorescens* and *B. megaterium*, therefore, they were used for management these fungi in plot experiment. In addition the fungus

F. solani was the most affected by the tested bioagents followed by *R. solani* then *M. phaseolina*.

Table1. Effect of the culture filtrate of five bacterial bioagents on the linear growth of *F.solaini* fungi, 4 days after incubation at $28\pm 2^{\circ}\text{C}$

Bioagents	Linear growth (mm) at concentration of (%)					Mean
	0.0	10	20	40	60	
B.megaterium	90.0	71.0	40.2	16.4	0.0	43.5
B.pumilus	90.0	78,8	45.6	24.8	11.3	50.1
B.subtilis	90.0	77.2	44.2	19.6	8.0	47.8
P.fluorescens	90.0	70.2	38.8	14.6	0.0	42.7
P.putida	90.0	72.0	41.8	18.0	0.0	44.4
Mean	90.0	74.2	42.1	18.7	3.9	-----

LSD at 5% for: Bioagents(B)= 2.3 , Concentration (C)= 3.2 and B x C= 3.7 .

Table2. Effect of the culture filtrates of five bacterial bioagents on the linear growth of *M.phasiolina* fungi, 4 days after incubation at $28\pm 2^{\circ}\text{C}$

Bioagents	Linear growth (mm) at concentration of (%)					Mean
	0.0	10	20	40	60	
B.megaterium	90.0	73.0	35.4	16.4	0.0	43.0
B.pumilus	90.0	82.4	53.8	37.0	26.6	58.0
B.subtilis	90.0	81.0	50.6	36.6	25.0	56.6
P.fluorescens	90.0	71.0	33.4	14.8	0.0	41.8
P.putida	90.0	72.2	34.0	17.4	0.0	42.7
Mean	90.0	75.9	41.4	24.4	10.3	-----

LSD at 5% for: Bioagents(B)= 2.6 , Concentration (C)= 3.5 and B x C= 3.9.

Table4. Effect of the culture filtrate of five bacterial bioagents on the linear growth of *R.solaini* fungi, 4 days after incubation at $28\pm 2^{\circ}\text{C}$

Bioagents	Linear growth (mm) at concentration of (%)					Mean
	0.0	10	20	40	60	
B.megaterium	90.0	70.0	33.2	11.4	0.0	40.9
B.pumilus	90.0	80.8	52.4	35.0	21.2	55.9
B.subtilis	90.0	75.8	41.6	20.2	10.4	47.6
P.fluorescens	90.0	68.2	31.0	9.8	0.0	39.8
P.putida	90.0	73.0	35.0	14.6	0.0	41.9
Mean	90.0	73.6	48.3	18.2	6.3	-----

LSD at 5% for: Bioagents(B)= 2.5 , Concentration (C)= 3.3 and B x C= 3.8.

Effect of Filtrate of Compost Tea on the Tinear Growth of the Tested Fungi

Table (4) reveals that the aquaus filtrate of compost caused significant inhibition to the linear growth of the three pathogenic fungi, four days after incubation at $28\pm 0^{\circ}\text{C}$ compared with

control treatment this inhibitory effect was gradually increased by increasing the concentration of the filtrate. Moreover, both *F.solani* and *R.solani* *M.phaseolina* failed to grow on the concentration of 60 % and *M.phaseolina* recorded 12.0 mm. linear growths.

Table4. Effect of filtrate of compost tea on the linear growth of the three tested fungi , four days after incubation at $28\pm 0^{\circ}\text{C}$.

Conc. (%)	Linear growth (mm) of			Mean
	<i>F. solani</i>	<i>M. phaseolina</i>	<i>R. solani</i>	
0.0	90.0	90.0	90.0	90.0
10	77.0	81.6	78.8	79.1
20	42.2	48.0	45.2	67.7
40	21.0	29.0	23.0	24.3
60	0.0	12.0	0.0	4.0
Mean	46.0	52.1	47.4	

LSD at 5% fot: Concentration (C) = 3.7, Linear growth (L) = 2.5 and B x C= 4.1.

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Effect of Soil Solarization on the Viability of the Three Pathogenic Fungi

The Average of the soil temperature at depths of 15 and 25 cm. was 59.7 and 51.3 °C for the mulched plots and 49.0 and 43.5 °C for the un-mulched plots during the period of soil solarization.

Table (5) shows that the pathogenic three fungi failed to re-isolate from the infected segments of solarized roots for 40 days at 15 and 25 cm.

Table5. Occurrence of the emerged fungi from the solarized segments of strawberry crowns and roots at the depth of 15 and 25 cm. 40 days after soil solarization

Depth (cm)	Kind of segments	No. of the emerged fungi					
		F. solani		M. phaseolina		R. solani	
		Un- mulched	Mulched	Un-mulched	Mulched	Un-mulched	Mulched
15	Crowns	6	2	10	5	8	3
	Roots	4	0.0	6	0.0	5	0.0
25	Crowns	9	4	12	7	10	6
	Roots	5	0.0	8	0.0	6	0.0

Effect of the Combination among the Bioagents B. Megaterium and P. Fluorescens, Compost, Soil Solarization on Management of Strawberry Crown and Root-Rot, Plant Growth Vigor, the Produced Fruit Yield and its T.S.S

Data shown in Table (6) show that the bioagents B. megaterium and P. fluorescens, compost and soil solarization resulted in significant reduction to strawberry crown and root-rot caused by the mixture of the inoculums of the three pathogenic fungi with improving the growth vigor of

depths. In addition, low frequency was recorded in case of solarized crown segments infected by F. solani, M. phaseolina and R. solani ,being 2, 5 and 3 at 25 depth and 4, 7 and 6 at 25 cm. depth, respectively. Meanwhile, the three fungi were isolated from the segments of control treatment (un-mulched soil) and the respective figures of the number of the isolated fungi from crown segments of control treatment (un-mulched soil) were 6, 10 and 8 at 15 depth and 9, 12 and 10 at 25 cm. depth.

strawberry plants. Also, significant increase to the produced fruits and it's total soluble solids (T.S.S.) were recorded when each of them was used alone or in their different combinations, compared with control treatment (infested with the causal fungi). In addition, no dead plants were occurred due to treating the infested soil with any of the tested treatments, each alone or in different combinations, as well as in case of control treatment of the un-infested soil compared with 54.2% dead plant in case of infested soil with tested fungi.

Table6. Effect of combination among compost , the bioagents B. megaterium and P. fluorescens and soil solarization on management of strawberry crown and root-rot diseases (Festival cv.) caused by three pathogenic fungi as well as plant growth vigor , fruit yield / plant and it's T.S.S., plot experiment.

Treatments	% Dead plants	% Crown and root-rot severity	Plant growth vigor	Ave. weight of fruits (g)/ plant	Total soluble Solid
Compost (C)	0.0	8.8	+++	364.7	16.3
B. subtilis (BS)	0.0	9.7	++	354.6	16.1
P. fluorescens (PF)	0.0	9.0	++	360.0	16.1
Soil solarization (SS)	0.0	10.2	++	358.0	16.0
C +BS	0.0	6.4	+++	385.0	16.4
C + PF	0.0	6.0	+++	388.5	16.4
C+SS	0.0	5.7	+++	383.0	16.4
BS + PF	0.0	6.0	+++	365.0	16.5
BS+SS	0.0	6.0	+++	365.0	16.5
PF+SS	0.0	6.0	+++	365.0	16.5
SS+C+BS+PF	0.0	0.0	++++	472.8	17.1
Control (Infested soil)	54.2	48.7	+	123.3	10.3
Control (Un-infested soil)	0.0	0.0	++++	475.0	17.2
L.S.D. at 5 %	5.7	4.5	----	6.4	0.8

Moreover, compost was more efficient in reducing the severity of infection by crown and

root-rot and improving plant growth vigor (very good growth) compared with the other three

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items of disease management. I.e. the bioagents *B. megaterium* and *P. fluorescens* and soil solarization when each of them was used alone, being 8.8, 9.7, 9.0 and 10.2%, respectively with moderate plant growth vigor. The combination between two the used items of disease management recorded low figures of disease severity compared with using each of them alone. Furthermore, excellent plant growth vigor and no apparent infection was detected when the combination among the bioagents *B. megaterium* and *P. fluorescens*, compost and soil solarization was used. Also, they produced fruit yield and T.S.S., to somewhat, similar to control treatment (un-infested with the causal fungi), being 472.8 and 475.0 g. fruit yield / plant 17.1 and 17.2 T.S.S, respectively. The highest disease severity, poor growth and the lowest fruit yield as well as T.S.S. Were noticed for strawberry plants grown in soil infested with the tested three fungi (infested with the causal fungus) , being 48.7% , (+) and 10.3 T.S.S..

DISCUSSION

Globally, the farmers are interested in reducing dependence on chemicals for growing the plants and those of pest management, especially in case of fruits and vegetable in order to avoid their residue in the produced yield. Therefore, disease management resort to use agriculture practices, sanitary methods, biological control, resistant cvs. , inducer resistance chemicals , nonparticles , plant extracts, soil solarization...ect to play important role in Integrated Pest Management (IPM) systems, especially in case of fruits and vegetables production. A model describing the several steps required for a successful IPM has been developed by Mc Spadden Gardener and Fravel (2002) . In this research, different items, compost, bacterial bioagents and soil solarization were evaluated for their efficacy in management of strawberry crown and root-rot, each alone or in different combination. The treatment with biopreparation induce systemic resistance as the main mechanism of activity on a plant or might be due to *P. fluorescens* produce different types of antibiotics including active 2, 4 diacetyl-phloroglucinole (2,4 DAPB), which control diseases and/or due to that *P. fluorescens* has several methods to control the disease such as production of antifungal compounds including siderophore production, nutrient competition and the induction of systemic resistance (Ramamoorthy *et al.*,2001). In addition, Meena *et al.* (2006) reported that the reduction in the infection by

plant pathogens and the increase in the plant length and fresh weight of the treated plants might be due to that *P. fluorescens* produces of indole acetic acid as a growth regulator as well as some antibiotic, *i.e.* pyrrolnitrin, pyoluterin and 2, 4 diacetyl phloroglucino. Bacillus-based as biological control agents (BCAs) have great potential in integrated pest management (IPM) systems; however, relatively little work has been published on integration with other IPM management tools (Jacobsen *et al.*, 2004).

Unfortunately, most research has focused on BCAs as alternatives to synthetic chemical fungicides or bactericides and not as part of an integrated management system. In this respect, in this work a combination among BCAs, compost and soil solarisation were used in managing strawberry crown and root-rot. Sterilized aqueous filtrate of the tested compost resulted in significant reduction to the linear growth of the tested fungi compared with control treatment. This reduction was gradually increased by increasing it's concentration. Using compost , the bioagents *B. megaterium* and *P. fluorescens* and soil solarization resulted in significant reduction to the severity of strawberry crown and root-rot diseases , improving plant growth vigor with significant increase to the fruit yield and it's total soluble solids (T.S.S.) compared with control treatment (infested soil). In addition, the combination between any of compost, the tested bioagents and soil solarization was more efficient in reducing disease severity, improving plant growth vigor and increasing fruit yield and it's T.S.S. than when each of them was used alone. Moreover, the combination among compost + the two bioagents *B. megaterium* and *P. fluorescens* + soil solarization was the most efficient in this respect, which no apparent infection by the disease was detected and the highest fruit yield and it's T.S.S. were obtained.

The highest efficiency of the combination between soil solarization and any of compost and *B. megaterium* or *P. fluorescens* may be greatly due to the drastic effect of solarization on the fungal propagules, which make them to be weak to resist the invasion by the tested bioagents. Also, compost can play a suitable medium for reproduction and establishment of the added bioagents and saprophytic microbes in the soil. Mehta *et al.* (2014) mentioned that compost plays great role in enhancing plant growth and reduces soil-borne plant diseases .It has been found that, the pathogenic three fungi, *i.e.* F.

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R. solani, *M. phaseolina* and *R. solani* failed to re-isolate from the infected segments of solarized roots for 40 days at 15 and 25 cm. depths. In addition low frequency of the emerged fungi from the infected segments of strawberry crowns by these fungi was recorded in case of solarized segments than control treatment. Meanwhile, the three fungi were isolated from the crown and root segments of control treatment (un-mulched soil).

Soil solarization is a special mulching process which causes hydrothermal disinfestation and other physical and biological changes in the soil which are beneficial to plant health and growth. In this regard, plastic sheets laid over moist soil during periods of high air temperature during summer, usually for 1–2 months, can greatly minimize and/ or eradicate great number of plant pathogens and pests as well as weeds.

The propagules of *F.o.f.sp. fragariae* were not detected at 5 cm. soil depth of the solarized soil and population at 10-15 depth showed a 60 % decrease (Kodama and Fuki, 1982 and Fahim et al., 1990). They added that as a result disease incidence was significantly reduced in outdoor cultivation of strawberry and in closed plastic houses population density of the pathogen fell sharply and remained low for 9 months following soil solarization.

The drastic effect of soil heating during summer is known and very important for lowering the population of the soil-borne fungi, especially those like the resistant fungal structures such as sclerotia and chlamydozoospores. The obtained data revealed that both *B. megaterium* and *P. fluorescens*, compost and soil solarization resulted in significant reduction to strawberry crown and root-rot caused by the mixture of the inoculums of the three pathogenic fungi with improving the growth vigor of strawberry plants. Also, significant increase to the produced fruits and its total soluble solids (T.S.S.) was recorded compared with control treatment (infested with the causal fungi).

In addition, no dead plants were occurred due to treating the infested soil with any of the tested treatments, each alone or in different combinations, as well as in case of control treatment of the un-infested soil compared with 54.2% dead plant in case of infested soil with tested fungi. Moreover, compost was more efficient in reducing the severity of infection by crown and root-rot and improving plant growth vigor (very good growth) compared with the

other three items of disease management. I.e. the bioagents *B. megaterium* and *P. fluorescens* and soil solarization when each of them was used alone. Furthermore, no apparent infection was detected in addition to excellent plant growth vigor, high fruit yield and its T.S.S when the combination among both bioagents, compost and soil solarization was used. Similar results were previously obtained by Mostafa et al. (1999); Abada et al. (2014) and Abada and Hassan (2017).

It is well known that IPM is a sustainable approach to managing pests by combining biological, cultural, physical and chemical tools in a way that minimizes economic, health and environmental risks. In theory, integration of several tools brings stability to disease management programs. Integration of bioagents with other disease management tools often provides broader crop adaptation and both more efficacious and consistent levels of disease control.

Furthermore, Noble and Coventry (2005) reported that composts have also been shown to suppress several diseases in the field, although the effects have been generally smaller and more variable than in container experiments. The disease suppressive effect of compost generally increased with rate of application. Compost inclusion rates of at least 20% (v/v) are normally required to consistently obtain a disease suppressive effect, particularly in peat-based media, but significant disease suppression has been found at lower inclusion rates in soil. Kwok (1987) reported that copiotrophic bacteria re-colonize composts most rapidly (24-48 h) after peak heating of compost.

He added that the predominant bioagents in this group include strains of *Bacillus*, *Pseudomonas* and *Pantoea* species. Also, Lookwood (1988) reported that edaphic microorganisms stimulated by compost amendments contribute to the suppressive activity of the amended soil through four control mechanisms, i.e. antibiosis, competition, predation hyperparasitism and the induction of systemic acquired resistance in the host plant.

It is supposed that *Bacillus* spp. could have diverse plant response involved in synthesis and accumulation of antimicrobial phytoalexins (Hammond-Kosack and Jones, 1996), induction of hypersensitive response (He et al., 1993), production of defense-related proteins (Yu, 1995), production of activated oxygen species

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(Baker et al., 1993). Modification of plant cell wall by deposition of callose (Veit et al., 2001) and production of siderophore (Yu et al., 2011), this type of resistance to diseases is named as induced systemic resistance (ISR) (Van Loon, 2007 and De Vleeschauwer et al., 2009).

In recent years, there has been a growing interest in BCAs bacteria due to their efficacy as bioagents in many crops (Akram et al.; 2013; Zaher et al., 2013 and Abada and Ahmed, 2014 and Abada and Hassan, 2017). Stapleton et al. (1985) reported that following soil solarization, growth of microflora beneficial to plant growth or antagonistic to pathogens and pests may slow the re-infestation of the soil by these organisms for more than one growing season.

Moreover, the availability of increased mineral nutrients following solarization may reduce crop fertilization requirements. It has been mentioned that phytopathologists have begun to characterize the determinants and pathways of induced resistance stimulated by bioagents and other non-pathogenic microbes (Bargabus, et al. 2004). The first of these pathways, termed systemic acquired resistance (SAR), is mediated by salicylic acid (SA), a compound which is frequently produced following pathogen infection and typically leads to the expression of pathogenesis-related (PR) proteins.

These PR proteins include a variety of enzymes some of which may act directly to lyse invading cells, reinforce cell wall boundaries to resist infections, or induce localized cell death. A second phenotype, first referred to as induced systemic resistance (ISR), is mediated by jasmonic acid (JA) and/or ethylene, which are produced following applications of some nonpathogenic rhizobacteria. Interestingly, the SA- and JA- dependent defense pathways can be mutually antagonistic, and some bacterial pathogens take advantage of this to overcome the SAR.

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