Bulletin of the National Research Centre

RESEARCH Open Access

Efficacy of soil solarization on black root rot disease and speculation on its leverage on nematodes and weeds of strawberry in Egypt

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Abstract

Background: Strawberry (*Fragaria ananassa* Duch.) is an economically important crop in Egypt. Yet complex black root rot disease of strawberry caused by *Fusarium solani*, *Rhizoctonia solani*, and *Pythium* sp. can cause considerable yield losses. Therefore, this study aimed at evaluating different aspects of soil solarization against this disease. Such an evaluation would better be viewed in the context of other beneficial effects of soil solarization on nematodes and weeds.

Materials/methods: Growth agar disks, growth suspension, and resting stages of strawberry black root rot fungi were evaluated at different temperatures and exposure times using digital hot water bath. Cloth bags artificially infested with single fungal species were buried into the soil before soil solarization at soil depths of 1–10, 11–20, and 21–30 cm at three spots of each plot for each of the abovementioned fungi for 3, 6, or 9 weeks. The disease incidence and severity in solarized and un-solarized soil was compared with the application of the fungicide Actamyl. Effects of soil solarization on nematodes and weeds were also consulted.

Results: The lethal temperature to *F. solani*, *Pythium* sp., and *R. solani* was 58, 58, and 56 °C, respectively when exposure time was 1 min. Chlamydospores were killed at 62 °C while sclerotia were killed at 58 °C in hot water for 1 min. Maximum soil temperature in solarized soil was raised by 15, 14, and 12 °C at depths of 1–10, 11–20, and 21–30 cm as compared with non-solarized soil. Solarization for 3, 6, and 9 weeks significantly reduced the disease incidence and severity and increased the strawberry yield. Complete reduction in total count of all tested fungi was obtained after 9 weeks at all tested depths. A review of collective soil pest and pathogen control via solarization documented its beneficial application.

Conclusion: The study may exploit hot months in Egypt for soil solarization against the serious root rot disease either singly or in an integrated pest management program.

Keywords: Black root rot, Plant pathogen control, Soil solarization, Strawberry

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Background

Strawberry (*Fragaria ananassa* Duch.) is globally one of the most economically important crops. It is grown under a wide range of climatic conditions as wild and cultivated plants producing small delicious fruits (Kurze et al. 2001). It is favored worldwide for the unique flavor and valuable macronutrients (Bianco et al. 2009). Yet strawberry black root rot ranks high among common diseases that can cause considerable yield losses in Egypt (El-Shemy et al. 2013). This disease is caused by several soil-borne fungal pathogens (Fang et al. 2012; Ahmed and El-Fiki 2017) such as *Fusarium oxysporum* (Juber et al. 2014), *Macrophomina phaseolina* (Hutton et al. 2013), *Pythium* spp. (Abdel-Sattar et al. 2008), *Phytophthora* spp. (Mingzhu 2011), and *Rhizoctonia* spp. (Fang et al. 2013). This complex disease is characterized by feeder rootlet killing, plant deterioration and blackening of the main root system as well as a decline in vigor and productivity of the plant stand causing a decrease in the crop yield (Abdel-Sattar et al. 2008; Fang et al. 2012; Ceja-Torres et al. 2014).

Soil solarization using transparent polyethylene plastic placed on moist soil during the hot months before planting usually increases soil temperature to levels lethal to many soil-borne plant pathogens, weeds, and nematodes (Primo and Cartia 2001; Abd-El-Kareem et al. 2004; Culman et al. 2006; Farag-Eman and Fotouh 2010). Hence, the present study was designed to test the effect of subjecting such fungal pathogens which cause the root rot disease to various categories of raised temperature under various forms, mainly solarization. However, being the only disease among other diseases and pests that is examined herein by solarization, such a study might be construed as imperfect from a collective soil pest and pathogen control or strawberry yield increase perspective. Therefore, in addition to evaluating the effect of soil solarization for different periods against strawberry black root rot and fruit yield (or for reducing populations of black root rot causing fungi in the soil, thus enhancing plant health), we briefly reviewed the beneficial effects of solarization on plant-parasitic nematodes (PPN) and weeds.

Materials and methods

Fungal pathogens and plant material

Pathogenic isolates of *Fusarium solani*, *Rhizoctonia solani*, and *Pythium* sp., the most obvious causal agents of black root disease of strawberry plants (Abd-El-Kareem et al., 2019), were kindly provided by Plant Pathology Department, National Research Centre, Giza, Egypt. Strawberry seedlings (cv. Festival) were obtained from Vegetable Crops Research Department, Agricultural Research Centre, Giza, Egypt.

Testing of hot-water treatments on viability of black root rot fungi

Viability of agar disks with mycelia and resting stages of *R. solani*, *F. solani*, and *Pythium sp.* was examined according to the method described by Whiting et al. (2001). Growth agar disks, growth suspension, and resting stages of strawberry black root rot fungi were evaluated at different temperatures and exposure times using digital hot water bath (Neslab GP-300 Series Constant Temperature Bath, Union City, CA). Screw-cap glass vials, 20 cm long and 20 mm in diameter, containing 20 ml sterilized water were placed in water path at different temperatures. Disks of agar with mycelia and spores, 6-mm diameter, were cut from the grown edge of 10-day-old cultures of *R. solani*, *F. solani*, and *Pythium* sp. growing on potato dextrose agar (PDA) medium. Agar disks were transferred to screw-cap glass vials, 20 cm long and 20 mm in diameter, containing 20 ml sterilized water placed in water path at 25, 50, 52, 54, 56, and 58 °C, for different exposures time, i.e., 1, 10, 20, and 30 min. These agar disks were then dried using sterilized filter paper and transferred into petri-plates containing PDA medium. Five screw-cap glass vials and 3 disks per each were used for each treatment. Viability of mycelia from agar disk that had been subjected to previous temperatures with different exposure times was assessed by planting treated disks on PDA medium and incubated at 25 °C for 5 days. Radial colony diameter was measured.

Preparation of fungal inocula

Inocula of *R. solani*, *F. solani*, and *Pythium* sp. were prepared by culturing each species on 50-ml potato dextrose broth (PDB) medium in 250-ml Erlenmeyer flasks for 15 days at 25–27 °C and each fungal inoculum was prepared as follows: inoculum of *F. solani* was prepared from the growing upper solid layers and colony forming units (cfu) were adjusted to 10^6 cfu/ml using hemocytometers' slide, where soil infestation was done at 50 ml/kg soil (Elad and Baker 1985). Inoculum *R. solani* was prepared from the growing upper solid layers which were washed and air-dried with sterilized filter paper layers. The air-dry mycelium was blended in distilled water to obtain inocula pieces of 1–2 mm in diameter. Soil infestation was done at 2 g dry mycelium/kg soil (Al-Mahareeq 2005). *Pythium* sp. inoculum was similarly prepared and propagules were adjusted to 10^6/ml. Soil infestation was done at 50 ml/kg soil (Lu et al. 2004).

Soil infestations with black root rot fungi

Sandy-loam soils were sterilized at 120 °C for 1 h. Sterilized soils were singly infested with inoculum of *R. solani*, *F. solani*, or *Pythium* sp. as mentioned before. Artificially infested soils were put into cloth bags at 1 kg infested soil/ bag.

Mulching soil with transparent polyethylene sheets

Three solarization periods, i.e., 3, 6, and 9 weeks were applied. Soil solarization started at the beginning of the last week of June 2017. All plots were irrigated to field capacity and 4 plots (3 m × 9 m) were covered with 200-μm thick transparent polyethylene sheets for 3, 6, or 9 weeks and then removed.

Buried cloth bags in infested soil

Cloth bags similarly infested with single species were buried into the soil before soil solarization at soil depths of 1–10, 11–20, and 21–30 cm at three spots of each plot for each of the abovementioned fungi and solarization periods.

Efficacy of soil solarization on soil temperatures

Average of maximum and minimum soil temperatures in solarized and non-solarized soil was recorded weekly using Thermo-couples during solarization period at soil depths of 1–10, 11–20, and 21–30 cm to estimate the thermal inactivation potential of solarization.

Quantification of black root rot fungi

Cloth bags infested with pathogenic fungi were buried in the soil before soil mulching as mentioned before. After removing the polyethylene sheets, the buried bags of each certain level in either solarized or un-solarized plots were collected. Total count of pathogenic fungi in solarized and un-solarized soil was compared with their count before soil mulching. Total count of pathogenic fungi was done according to Porras et al. (2007). The fungal colonies per gram of dry soil were calculated. The resulting reduction was counted as follows:

% Reduction = [(No. of colonies in control–No. of colonies in treatment)/No. of colonies in control] × 100

Field experiment

Field plots (3 m × 8 m) at El-Qalioubia governorate, Egypt, were irrigated to field capacity and 4 plots were similarly covered with 200-μm thick transparent polyethylene sheets. The field was highly infested with strawberry black root fungi. Experiments were conducted under natural-infested soil in plots each comprised of 8 rows (32 holes/row and one seedling were sown in each hole) in a randomized complete block design with three replicates (plots) for each treatment. Strawberry seedlings were transplanted in well-drained loamy clay soil to a depth of 10 cm in October 2017 and uprooted at the end of May 2018. Irrigation and nutrients such as phosphorus, nitrogen, and potassium were added as recommended (El-Shemy et al. 2013).

Treatments

Solarization periods of 3, 6, and 9 weeks were tested to study their effect on strawberry black root rot and yield relative to the control (un-solarized soil) under field conditions. Solarization started at the beginning of the last week of June 2017. Actamyl (3 g/L) fungicide (active ingredient: thiophanate-methyl) was applied for comparison.

Disease incidence and severity and yield estimate

Disease incidence was calculated 100 days after transplanting as follows:

% disease incidence = (number of infected plants/total number of plants) × 100

Disease severity was similarly recorded according to Morocko (2006) as follows:

0 = plant well developed, no disease symptoms; 1 = no visible symptoms on above-ground parts, 25% of roots discolored; 2 = plant slightly stunted, black necrosis on petiole bases, 26–50% of roots discolored; 3 = plant stunted, black necrosis on petiole bases, yellowing and death of outer leaves, 51–75% of roots discolored; 4 = plant severely stunted, outer leaves collapsed, younger leaves bluish green and wilting, > 75% of roots discolored; 5 = plant dead.

$$\text{Disease severity}\% = \frac{\Sigma\ (\text{Disease grade} \times \text{number of plants in each grade})}{\text{Total number of plants} \times \text{highest disease grade}} \times 100$$

Accumulated strawberry yield (tons/feddan) for each treatment was determined. Statistical analysis: Tukey test for multiple comparisons among means was utilized (Neler et al. 1985).

Results

Hot-water treatments

Viability of agar disks with mycelia and resting stages of *R. solani*, *F. solani*, and *Pythium* sp. was tested to determine the lethal temperature of the pathogenic fungi. Results in Table 1 reveal that increasing exposure time and/or temperature could block growth of the tested

Table 1 Viability of mycelia agar disks of strawberry black root rot fungi as affected by hot water at different temperatures and exposure times

Hot water (°C)	Exposure time (minutes)											
	F. solani				*Pythium* sp				*R. solani*			
	1	10	20	30	1	10	20	30	1	10	20	30
25	+	+	+	+	+	+	+	+	+	+	+	+
50	+	+	+	+	+	+	+	+	+	+	+	+
52	+	+	+	+	+	+	+	+	+	+	+	+
54	+	+	+	+	+	+	+	+	+	+	–	–
56	+	–	–	–	+	+	–	–	–	–	–	–
58	–	–	–	–	–	–	–	–	–	–	–	–

(+) Indicate growth, (–) indicate no growth

fungi. The lethal temperatures to *F. solani*, *Pythium* sp., and *R. solani* were 58, 58, and 56 °C respectively at 1 min of exposure time.

Effect of different exposure times and temperatures on resting stage viability

Results in Table 2 indicate the effect of different exposure times and temperatures on resting stage germination of strawberry root rot fungi under laboratory conditions. Chlamydospores were more resistant to high temperature as they were killed at 62 °C but sclerotia were killed at 58 for *R. solani* when exposed to hot water for one minute.

Effect of soil solarization on soil temperatures

Averages of maximum and minimum soil temperatures in solarized and non-solarized soil were recorded. Results in Table 3 indicate that maximum temperatures in solarized soil increased by 15, 14, and 12 °C at depths of 1–10, 11–20, and 21–30 cm of soil surface as compared with un-solarized soil. The highest temperatures were 56, 52, and 46 °C at the three depths, respectively.

Effect on total count of black root rot fungi in soil

Results in Table 4 indicate that fungal populations decreased in both mulched and un-mulched soils at the end of the experiment. However, solarization for 6 and 9 weeks was more effective in reducing the pathogens population. Complete reduction in total count of the tested fungi was obtained after 9 weeks at the three solarized depths. Meanwhile, solarization for 6 weeks caused a complete reduction in the total count only at a depth of 1–10 and 11–20 cm.

Effect of solarization on strawberry black root rot disease

Results in Table 5 show that all tested solarization periods significantly reduced the strawberry black root rod disease incidence and severity. The most effective solarization periods were 6 and 9 weeks which reduced the disease incidence and severity by 87.4, 84.6, 88.4, and 87.2%, respectively. Solarization for 3 weeks had a moderate effect.

Table 2 Resting stage viability of strawberry black rot fungi as affected by hot water temperatures and exposure times

Hot water (°C)	Exposure time							
	Chlamydospores				Sclerotia			
	F. solani				*R. solani*			
	1	10	20	30	1	10	20	30
25	+	+	+	+	+	+	+	+
52	+	+	+	+	+	+	+	+
54	+	+	+	+	+	+	–	–
56	+	+	+	+	+	+	–	–
58	+	+	+	+	–	–	–	–
60	+	+	+	–	–	–	–	–
62	–	–	–	–	–	–	–	–

(+) Indicate growth, (–) indicate no growth

Table 3 Effect of soil solarization on soil temperatures

Soil category	Depth	Maximum temperatures (°C)	Minimum temperatures (°C)
Solarized soil	1–10	56 a	38 a
	11–20	52 b	35 b
	21–30	46 c	33 c
Un-solarized soil	1–10	41 d	27 d
	11–20	38 e	24 e
	21–30	34 e	21 f

Figures with the same letter in a column are not significantly different ($P \leq 0.05$) using Tukey test

Effect of solarization on strawberry yield

Results in Table 6 show that all tested solarization periods significantly increased the strawberry yield. The most effective solarization periods were 6 and 9 weeks which increased the yield by 107.7 and 115.4%, respectively. Solarization for 3 weeks had moderate effect.

Discussions

Soil solarization proved effective for healthy strawberry cultivation in terms of reducing strawberry root rot disease incidence and severity as well as increasing strawberry fruit yield under different aspects in the current study. These included three different species of pathogenic fungi each at three soil depths. The present results are in agreement with those of Katan et al. (1976) and Katan (1980) who demonstrated that the population of soil-borne fungi *F. oxysporum*, *R. solani*, and *Sclerotium rolfsii* were reduced by 62 to 100% at 5 to 25-cm depths in solarized soil. Reduction in disease incidence/severity and increase in obtained yield due to soil solarization were reported by many investigators (Katan 1980; Osman et al. 1986; Abd-El-Kareem et al. 2004; Culman et al. 2006; Farag-Eman and Fotouh 2010). Pullman et al. (1981) presented a detailed study on thermal death of three soil-borne plant pathogens as affected by time and temperature of the treatment. They found that the inability of organisms to tolerate high temperatures is related to an upper limit in the degree of fluidity of membranes beyond which breakdown of membrane function may be associated with membrane instability and the sustained inactivation of respiratory enzymes (Sundarum 1986). Also, changes occur in the structure of the soil, insoluble mineral substances available for plant and microbial growth, and in the populations of soil-borne microorganisms during heat treatments of soil (Chen and Katan 1980; Stapleton and DeVay 1984;

Table 4 Percentage reduction in pathogenic fungi at three soil depths as affected with different periods of solarized and un-solarized soil under field conditions

Treatments	Solarized periods (weeks)	Soil depths (cm)	Reduction in black root rot fungi %		
			F. solani	*R. solani*	*Pythium* sp.
Solarized soil	3	1–10	72	75	73
		11–20	62	66	64
		21–30	45	51	45
	6	1–10	100	100	100
		11–20	100	100	100
		21–30	82	100	86
	9	1–10	100	100	100
		11–20	100	100	100
		21–30	100	100	100
Un-solarized soil	3	1–10	20	22	23
		11–20	10	11	9.5
		21–30	8	11	8
	6	1–10	40	46	41
		11–20	24	32	21
		21–30	10	22	13
	9	1–10	65	68	66
		11–20	48	52	44
		21–30	32	35	33

Figures with the same letter in a column are not significantly different ($P \leq 0.05$) using Tukey test

Stapleton et al. 1985). These changes that affect the inoculum density of plant pathogens and their aggressiveness and survival are probably not limited to the rot disease. Moreover, crop yield increase due to solarization is not exclusively and definitely attributed to the control of pathogens and weeds but rather to a combination of mechanisms. Plant growth increased in solarized soil without pests and pathogens (Stapleton and DeVay 1984). It could be that immeasurable or undetectable pathogens and pests such as PPN are also controlled. Also, solarization may increase some soluble nutrients such as calcium, nitrogen, and magnesium and make them more available to plants in solarized soil. A third assumption is that beneficial microorganisms such as actinomycetes, mycorrhizal fungi, and species of *Trichoderma* can survive the solarization process or recolonize the soil rapidly (Stapleton et al. 1985; Kishore et al. 2008).

In this context, soil temperatures increase to lethal levels for many other soil-borne plant pathogens such as PPN and weeds especially during summer months (Elmore et al. 1997; Primo and Cartia 2001; Abd-El-Kareem et al. 2004; Culman et al. 2006; Farag-Eman and Fotouh 2010). Hence, the expected increase in temperature with climate change can reinforce the expansion of other alternatives to methyl bromide rather than programs in Egypt that are mostly limited to the evaluation of chemical treatment and biocontrol agents.

Table 5 Black root rot disease of strawberry plants as affected with different periods of solarized and un-solarized soil under field conditions

Treatments	Solarized periods (weeks)	Black root rot disease			
		Disease incidence	Reduction %	Disease severity	Reduction %
Solarized soil	3	18.0 b	58.1	12.0 b	69.2
	6	5.4 c	87.4	6.0 c	84.6
	9	5.0 c	88.4	5.0 c	87.2
Actamyl 3 g/L	0.0	8.0	81.4	9.0	76.9
Un-solarized soil	9	43.0 a	0.0	39.0 a	0.0

Figures with the same letter in a column are not significantly different ($P \leq 0.05$) using Tukey test

Table 6 Strawberry yield as affected by different periods of solarized and un-solarized soil under field conditions

Treatments	Solarization (in weeks)	Strawberry yield	
		Yield (tons/feddan)	Increase %
Solarized soil	3	9.5 b	46.2
	6	13.5 a	107.7
	9	14.0 a	115.4
Actamyl 3 g/L	0	9.5 b	46.2
Un-solarized soil	0	6.5 c	0.0

Figures with the same letter in a column are not significantly different ($P \leq 0.05$) using Tukey test

Both progressive phasing out of available chemical nematicides and expected heavier root-knot nematode infestations with warmer climate change should further materialize solarization as an imperative component in integrated pest management (IPM). Candido et al. (2008) found that yearly solarization is needed for root-knot nematodes (RKN) control of greenhouse tomato and melon as very susceptible hosts but two- or three-yearly applications can effectively control weeds. Raising thermal-efficiency films or integration with other PPN control approach can extend the residual impacts of solarization concerning RKN on strawberry. Two-consecutive year solarization had greatly suppressed RKN and increased yield of tomato and melon (Candido et al. 2008). Samtani et al. (2012) found that steam + solarization treatments were so effective in weed control that both together were similar to combined fumigation.

Generally, plasticulture technologies for strawberry cultivation proved effective against PPNs in Egypt (Mahdy and Midan 2011; Abd-Elgawad 2019). When colored polyethylene sheets (transparent, red, black, green, and blue) were used in such a technology, the best reduction of RKN occurred in transparent sheet relative to the others (Bakr et al. 2013). Plasticulture considerably reduced population levels of numerous PPN species (Sauerborn et al. 1990).

Conclusion

Under the warm weather conditions of Egypt and expected increase in temperature with potential climate change, solarization is an effective and safe alternative for methyl bromide and other banned chemicals for the management of strawberry pests and diseases. Its reduction of strawberry root rot disease incidence and severity, as well as increase strawberry fruit yield collectively with its control of other pests and pathogens reported in the abovementioned references can offer a satisfactory tool for IPM programs of strawberry; so further feasible studies of such programs are warranted.

Abbreviations

cfu: Colony forming units; IPM: Integrated pest management; PPN: Plant-parasitic nematodes; PDA: Potato dextrose agar; PDB: Potato dextrose broth

Acknowledgements

This study was supported in part by the US-Egypt Project cycle 17 (no. 172) entitled "Preparing and evaluating IPM tactics for increasing strawberry and citrus production."

Authors' contributions

All authors participated in the development, implementation, and subsequently writing of the research plan. All authors read and approved the final manuscript.

Funding

Financial support made by US-Egypt Project fund for Project cycle 17 (no. 172) and National Research Centre, Egypt, is gratefully acknowledged.

Availability of data and materials

The datasets used and/or analyzed during the current study are available from the corresponding author on reasonable request.

Ethics approval and consent to participate

Not applicable.

Consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

Received: 31 July 2019 Accepted: 19 November 2019
Published online: 05 December 2019

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