Assiut J. Agric. Sci., (47) No. (6-1) 2016 (87-99) *ISSN: 1110-0486*
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Suppression Effect of Vermicompost Tea on Onion White Rot

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Received on: 8/8/2016 **Accepted for publication on:** 17/8/2016

Abstract

Vermicompost tea (VCT) applications at the rate of 5ml/pot three or four times with irrigation water gave the best reduction of onion white rot with 30.0% infection, while the fungicide Folicure gave 10.0% infection compared with 70.0% infection at non-treated plants. In field experiments all tested treatments reduced the percentage of infection compared with non-treated plants. The best treatment in reducing onion white rot was VCT at the rate of 15 L/fed four times with irrigation water that gave 20.0% infection, while the fungicide Folicure gave 8.3% infection. On other hand, VCT treatment at the rate of 15L/fed four times with irrigation water increased onion yield, plant height, no. of leaves/plant, onion fresh and dry weight more than all other treatments even Folicure treatment. Also, all VCT treatments increased total count of soil bacteria, fungi and actinomycetes Compered with control.

Keywords: *Onion white rot, Sclerotium cepivorum and vermicompost tea.*

Introduction

The soil-borne fungus, *Sclerotium cepivorum* Berk. is the causal agent of Allium white rot (Entwistle, 1990a) and one of the most serious diseases of onions (*Allium cepa* L.) (Merriman *et al.*, 1980 and Metcalf and Wilson, 1999). The disease can reduce yields to uneconomic levels in 4 years of successive onion crops (Coley-Smith, 1987). The pathogen persists in the soil in the absence of host plants as sclerotia and can survive in this form for more than 20 years (Coley-Smith and Parfitt, 1986 and Coley-Smith *et al.*, 1987). Following their production, sclerotia are constitutively dormant for 1-3 months and will then only germinate in the presence of host plants (Coley-Smith *et al.*, 1987).

Land free of Allium white rot in onion-growing areas is decreasing, forcing onion production into structurally poorer soils (Entwistle, 1990b). Control of white rot and the return of infested land to onion production are clearly major concerns of the onion-growing industry, particularly with the loss of methyl bromide as a soil sterilant (Merriman *et al.*, 1980 and De Ceuster and Hoitink, 1999). A numbers of control methods have been researched including fungicide application (Davies and Savinelli, 1994 and Stewart *et al.*, 1994), soil fumigants (Entwistle, 1990a), soil solarisation (Porter and Merriman, 1983 and Melero-Vara *et al.*, 2000), use of biological control agents (Stewart *et al.*, 1994 and Gerlagh *et al.*, 1996) and germination stimulants (Coley-Smith and Parfitt, 1986). Levels of disease control vary according to the method used and the time of year (Coley-Smith and Parfitt, 1986 and Melero-Vara *et al.*, 2000), with no one method offering complete control.

Hand *et al.*, (1988) defined Vermicompost (VC) as a low cost technology system for processing or treatment of organic waste. Vermicompost aqueous extracts are much easier to handle and apply than solid VC, which are bulky and heavy (Yardim *et al*., 2006).

The effect of compost and compost tea on suppression of certain plant diseases have been discussed by multiple studies (Scheuerell and Mahaffee, 2004, Scheuerell *et al*., 2005, Choi *et al*., 2007 and Zmora Nahum *et al*., 2008). However, relatively little work has been done to investigate the effect of vermicompost teas (VCTs) on yield and nutritional quality of vegetable crops. However, application of VCTs has been shown to improve plant health, crop yield, and nutritive quality (Gamaley *et al*., 2001 and Pant *et al*., 2009). Welke (2005) showed that both aerated and non-aerated compost tea extracted from composted animal manure have similar positive effects on straw-berry yield and suppression of *Botrytis cinerea*. Edwards *et al*. (2007) demonstrated significant suppression of plant parasitic nematodes, aboveground foliar arthropod pests and both foliar and root plant diseases by soil drenches of aqueous extracts produced from VCs.

The objectives of this study were to determine the effect of VCT on white rot of onion incidence under greenhouse and field conditions and to more elucidate their impacts on bulb yield and total count of soil bacteria, fungi and actinomycetes compared with control.

Materials and Methods

Sclerotium cepivorum

An isolate of *S. cepivorum* was isolated from infected onion plants collected from Menia Governorate. Identified based on the morphological characteristics as mentioned by Mordue (1976). The isolate was used to inoculate sterilized barley seeds medium for 3 weeks at 20^{o} C (Van der Meer *et al*., 1983) and used as inoculum.

Vermicompost tea

VCT was obtained from Central Laboratory for Agricultural Climate, ARC, Giza, Egypt. VC was mixed with tap water as 1:10 (w/v) in 19 L plastic bucket. The mixture was left open for 7 days at 20-21 ^{o}C and stirred once on day 4 of preparation. The resulted VCT was filtered through a nylon membrane just before application and stored at 4^{o} C (Weltzein, 1991).

The Fungicide Folicure

The fungicide Folicure 25% EC (tebuconazole 25%) used in this investigation as a comparison with the treatments was obtained from Ministry of Agriculture, Egypt.

Greenhouse experiments

Pot experiments were carried out in the greenhouse of Onion, Garlic and Oil Crops, Plant Path. Res. Inst., ARC during 2014/2015 season to evaluate onion white rot incidence under different treatments of VCT.

Plastic pots (25-cm-diam.) filled with sterilized sand-clay soil (1:1 v/v) were infested with the prepared *S. cepivorum* inoculum at the rate of 2% w/w, 7 days before transplanting. Four pots were used for each treatment and control (infested pots free treatment). Five seedlings of Giza 6 cv. (60-day-old) were transplanted in each pot 1st November and irrigated when needed.

Vermicompost tea treatment

Vermicompost tea was added to the soil with irrigation water at the rate of 1, 2, 3, 4 and 5 ml/pot. for each concentration were used i.e., after transplantation; after transplantation and one month later; after transplantation, one and two months later; after transplantation, one, two and three months later.

Folicure treatment

Transplants were dipped for 5 min. in the Folicure 25% (25 ml/L water) just before transplanting then the grown plants were sprayed at 6 and 12 weeks after transplantation (187.5 ml/100L water).

Field experiments

Field experiments were carried out during 2014/2015 and 2015/2016 seasons in naturally infested soil with *S. cepivorum* at Mallawy Agric. Res. St., Menia Governorate. Sixty days old, onion transplants (Giza 6 cv.) ware transplanted in both seasons was in 1st November. Experiments were designed as complete randomized blocks. Four replicate plots were used for each treatment and control. The area for each plot was 10.5 m^2 (3.0 X 3.5 m). All treatments received the same normal agricultural practice until harvest at April.

Vermicompost tea treatment

VCT was added to the soil with irrigation water at the rate of 3, 6, 9, 12 and 15 L/fed. Four treatments for each concentration were used i.e., after transplantation; after transplantation and one month later; after transplantation, one and two months later; after transplantation, one, two and three months later.

Folicure treatment

Seedlings were dipped for 5 min. in the Folicure 25% (25 ml/L water) just before transplanting then grown plants were sprayed at 6 and 12 weeks after transplantation (187.5 ml/100L water).

Disease index and estimated parameters

At the end of each season under greenhouse and open field, percentage of infection

$$\frac{\text{treatment} - \text{control}}{\text{control}} X100 \quad \text{and efficacy} \quad \frac{\text{control} - \text{treatment}}{\text{control}} \; x100$$

were assessment, while onion bulb yield was estimated at harvest of each season under open field conditions.

Total count of soil bacteria, fungi and actinomycetes

Estimation of soil bacteria, fungi and actinomycetes in rhizosphere region were made after two weeks from each treatment using the dilution plate technique (Warcup, 1951). Soil extract agar medium for bacteria, potato dextrose rose bengal agar medium for fungi and oat meal agar medium for actinomycetes were used. The inoculated plates were incubated at 27 ^oC for two days for bacteria, three days for actinomycetes and four days for fungi. Colony forming units (cfu) were counted by using a colony counter

Statistical analysis

The data were statistically analyzed and significance among means was assessed by least significant difference (LSD) at 5% probability level using SAS ANOVA program V.9 (Anonymous, 2014).

Results

The data present in Table (1) show that, VCT applications at the rate of 5ml/pot three or four times with irrigation water gave the best reduction of onion white rot with 30.0% infection and 57.14% efficacy compared with non-treated plants. There is an inverse relationship between the concentration of vermicompost tea and percentage of infection, as well as between the times of application and percentage of infection. Treatments at the rate of 1, 2 and 3ml/pot gave no significant different on disease infection compered with control. Meanwhile, the fungicide Folicure as check control, gave the best reduction of infection (10.0 % infection and 85.71 % efficacy) compared with control.

Table 1. Effect of vermicompost tea and fungicide Folicure (25%) on white rot infection of onion plants cv. Giza 6 under greenhouse conditions, during growing seasons 2014/2015.

Treatment	Application times*	Infection%	Efficacy%
1 ml/pot	1	75.0	-7.14
	2	75.0	-7.14
	3	70.0	0.00
	4	75.0	-7.14
2 ml/pot	1	75.0	-7.14
	2	70.0	0.00
	3	65.0	7.14
	4	65.0	7.14
3 ml/pot	1	70.0	0.00
	2	65.0	7.14
	3	65.0	7.14
	4	60.0	14.29
4 ml/pot	1	75.0	-7.14
	2	50.0	28.57
	3	50.0	28.57
	4	45.0	35.71
5 ml/pot	1	50.0	28.57
	2	40.0	42.86
	3	30.0	57.14
	4	30.0	57.14
Folicur		10.0	85.71
Control		70.0	-
LSD 0.05		14.54	-

* Vermicompost tea were added with irrigation water as follow: (1) after transplantation; (2) after transplantation and one month later; (3) after transplantation, one and two months later; (4) after transplantation, one, two and three months later.

According to presented data in Table (2) show that all treatments reduced the percentage of infection compared with non-treated plants (63.5% infection). Treatments efficacy increased gradually with increase of treatments volume. Also, increasing of application times decrease the percentage of infection under all tested concentrations. Treatments efficiency ranged between 68.5% with VCT at the rate of 15 L/fed four times with irrigation water and 1.6% with VCT at the rate of 3L/fed one time with irrigation water. The best treatment in reduction onion white rot was VCT at the rate of 15L/fed four times with irrigation water that gave 20.0% infection and 68.5% efficacy. From other hand, the check control (Folicure) gave 8.3% infection and 86.9% efficacy compared with non-treated plants.

Table 2. Effect of vermicompost tea and fungicide Folicure (25%) on white rot infection of onion plants cv. Giza 6 under naturally infested soil with *S. cepivorum* during growing seasons 2014/2015 and 2015/2016.

Treatment	Application times*	infection%		Mean	Efficacy%
		2014/15	2015/16		
3 L/fed	1	62.00	63.00	62.50	1.60
	2	60.40	61.50	61.00	4.00
	3	60.90	61.50	61.20	3.60
	4	62.00	62.50	62.30	2.00
6 L/fed	1	59.40	59.90	59.70	6.10
	2	58.30	58.80	58.60	7.80
	3	56.70	56.20	56.50	11.10
	4	57.30	54.80	56.10	11.70
9 L/fed	1	50.50	51.10	50.80	20.00
	2	51.10	49.10	50.10	21.10
	3	47.00	46.50	46.80	26.40
	4	43.80	42.70	43.30	31.90
12 L/fed	1	51.00	50.00	50.50	20.50
	2	38.00	38.50	38.30	39.80
	3	32.30	31.70	32.00	49.60
	4	31.20	32.30	31.80	50.00
15 L/fed	1	23.70	23.90	23.80	62.50
	2	23.20	23.50	23.40	63.20
	3	21.90	21.30	21.60	66.00
	4	19.80	20.30	20.10	68.40
Folicur		7.30	9.40	8.40	86.90
Control		63.00	64.00	63.50	-
LSD 0.05		1.65	1.67	-	-

* Vermicompost tea were added with irrigation water as follow: (1) after transplantation; (2) after transplantation and one month later; (3) after transplantation, one and two months later; (4) after transplantation, one, two and three months later.

According to data presented in all applications increased onion yield more than control unless treatment at the rate of 3L/fed two times that gave the same yield as control (7.0 kg/plot). Treatments with low concentrations i.e., 3 and 6 L/fed have no clear relation between application times and resulted onion yield. While there is a positive relationship between concentrations or application times and onion yield with 9, 12 and 15 L/fed. VCT treatment at the rate of 15L/fed four times with irrigation water increased onion yield more than all other treatments even Folicure treatment, which it gave 12.0 kg/plot with 72.1% increase, while Folicure treatment gave 10.3kg/plot with 47.1% increase in yield respectively compared with non-treated plants.

Table 3. Effect of vermicompost tea and fungicide Folicure (25%) on bulbs yield of onion under naturally infested soil with *S. cepivorum* at Mallawi during growing seasons 2014/2015 and 2015/2016.

Treatment	Application times*	Yield (kg/plot)		Mean	Increase (%)
		2014/15	2015/16		
3 L/fed	1	7.50	7.60	7.50	7.90
	2	7.30	6.70	7.00	0.00
	3	7.20	7.40	7.30	4.30
	4	7.70	7.70	7.70	10.00
6 L/fed	1	7.50	7.60	7.50	7.90
	2	7.70	7.60	7.60	9.30
	3	7.30	7.50	7.40	5.70
	4	8.10	8.20	8.20	16.40
9 L/fed	1	7.50	7.60	7.50	7.90
	2	8.10	8.50	8.30	18.60
	3	8.20	8.50	8.40	19.30
	4	8.60	8.70	8.60	23.60
12 L/fed	1	8.00	8.30	8.20	16.40
	2	8.90	9.20	9.00	29.30
	3	8.70	8.80	8.80	25.00
	4	9.20	9.10	9.20	30.70
15 L/fed	1	8.10	8.10	8.10	15.70
	2	8.70	9.00	8.90	26.40
	3	9.60	9.90	9.80	39.30
	4	12.00	12.10	12.00	72.10
Folicur		10.30	10.30	10.30	47.10
Control		7.20	6.80	7.00	-
LSD0.05		0.56	0.70	-	-

* Vermicompost tea were added with irrigation water as follow: (1) after transplantation; (2) after transplantation and one month later; (3) after transplantation, one and two months later; (4) after transplantation, one, two and three months later.

As show in Table (4), all treatments increased the total count of soil microorganisms, while Folicure treatment decreased the total count of soil microorganisms compared with non-treated plants in rhizosphere region. The total count of soil bacteria, fungi and actinomycetes increased positively with either increasing of treatment volume or application times. VCT at the rate of 15L/fed four times with irrigation water gave the highest count of soil bacteria, fungi and actinomycetes more than all other treatments and control.

Generally, the present results show that the application by VCT at the of 15L/fed four times with irrigation water can significantly control onion white rot and increased and bulbs yield compared to non-treated plants.

Table 4. Effect of vermicompost tea and fungicide Folicure (25%) on total count of soil bacteria, fungi and actinomycetes after two weeks from each treatment under naturally infested soil with *S. cepivorum* during growing seasons 2015/2016.

Treatment	Application times*	Total count (CFU)**		
		colony of Bacteria (10^5/g^{-1})	colony of fungi (10^6/g^{-1})	colony of actinomycetes (10^5/g^{-1})
3 L/fed	1	10.50	10.00	7.50
	2	11.00	10.50	8.00
	3	11.90	11.20	8.10
	4	12.52	11.70	9.00
6 L/fed	1	11.00	10.20	7.80
	2	12.20	10.92	8.89
	3	12.90	11.50	9.50
	4	13.59	11.90	10.60
9 L/fed	1	12.00	11.00	8.20
	2	13.12	11.48	9.60
	3	14.20	12.00	10.70
	4	16.60	13.10	11.64
12 L/fed	1	14.5	12.50	9.82
	2	16.00	13.04	10.43
	3	18.35	14.30	11.60
	4	20.00	14.95	12.22
15 L/fed	1	15.70	13.00	10.00
	2	18.00	13.70	10.50
	3	20.70	15.90	11.20
	4	23.70	17.50	13.90
Folicur		7.60	6.00	4.50
Control		10.20	9.0	5.80

*Vermicompost tea were added with irrigation water as follow: (1) after transplantation; (2) after transplantation and one month later; (3) after transplantation, one and two months later; (4) after transplantation, one, two and three months later. **CFU is colony-forming units

Discussion

Several studies have shown a positive effect of VCTs on suppression certain plant diseases such as *Botrytis* on green beans, strawberries, grapes and geraniums, leaf spot on tomatoes, bacterial speck in *Arabidopsis* and powdery mildew on apples (Hoitink *et al*., 1997; Zhang *et al*., 1998 and Al-Dahmani *et al*., 2003). Application of VCTs has been shown to improve plant health, crop yield and nutritive quality (Gamaley *et al*., 2001 and Pant *et al*., 2009). Results reported herein showed that VCT treatments reduced onion white rot incidence either in greenhouse or field conditions. Such results agreement with Scheuerell and Mahaffee (2004) who found that VCT suppressed the growth of *Pythium ultimum* under greenhouse conditions in a soilless media. Also Arancon *et al*. (2007) stated that VCT control or prevent various types of pathogenic fungi and diseases include *pythium, phytopthora, rhizoctonia, plectospora, verticillium,* powdery and downy mildews, bacterial blights, leaf spots, apple scab and grey mold. The treatment of VCT has been shown to reduce disease by necrotrophs as well as biotrophs (AL-Dahmani *et al*., 2003), and significantly decreased soil borne pathogens and various pests (Subler *et al*., 1998 and Sobha *et al*., 2003). More recent research has demonstrated significant suppression of plant parasitic nematodes, above-ground foliar arthropod pests and both foliar and root plant diseases by soil drenches of aqueous extracts, produced from VC (Edwards *et al*. 2007).

Vermicompost have much greater microbial biodiversity and activity than conventional thermophilic composts (Edwards and Arancon, 2004 and Pant *et al*., 2011). Aqueous extracts of vermicompost increased microbial populations and enhancing beneficial microbial communities and their effects on agricultural soils and plants (Arancon *et al*., 2007). If a broad range of microorganisms from VC pass to teas, their applications into a disease-infested planting medium or soil could suppress pathogen attacks, the suppression was clearly microbial since suppression properties of VC were lost after sterilization of the VC (Edwards *et al*., 2006). The factors involved in disease suppression have focused largely on comparative studies of the bulk properties of suppressive and non suppressive composts (Bonanomi *et al*., 2010). A consistent pattern that has emerged from these studies is the central role of microbial activity in compost-induced disease suppression (Hadar and Papadopoulou, 2012). The subset of bacteria that colonize seed shortly after sowing may be the most critical for disease suppression (Chen and Nelson, 2008 and Chen *et al*., 2012) because of their direct interference with pathogenesis in the infection court (Chen and Nelson, 2012). From other hand VCTs application can induce plants systemic resistance to make physiological changes that decrease its vulnerability to infection (Pieterse *et al*., 2003).

During vermicomposting the nutrients are released and converted into soluble and available forms to plants (Ndegwa and Thompson, 2001). Soluble mineral nutrients extracted have a positive effect on plant growth and yield with foliar and soil applications (Ingham, 2003 and Pant *et al*., 2009). VCTs as a foliar spray or soil drench have been demonstrated to improve plant health, yield and nutritive quality by enhancing beneficial microbial communities and their effects on agricultural soils and plants, improving the mineral nutrient status of plants and inducing the production of plant defense compounds that have beneficial bioactivities in humans (Weltzein. 1991, Hoitink, 1997. Diver, 2001, Scheuerell and Mahaffee, 2002 and Carpenter-Boggs, 2005). Certain groups of rhizobacteria found in VC and VCTs have been termed plant growth-promoting rhizobacteria (PGPR) they aggressively colonize plant roots and promote plant growth (Allison and Janice, 2006). It is also postulated that the action of living micro-organisms and microbial metabolites will stimulate plant growth (Diver, 2001 and Carpenter-Boggs, 2005). Increased root initiation, root biomass and shoot biomass are common effects of the application of VC and VCT and effects seen by application of synthetic hormones to plants (Tomati *et al.*, 1988). Growth regulators or phytohormones extracted from VC may also have a positive effect on initial root development and plant growth (Keeling, 2003 and Edwards 2006). The interaction between earthworms and microorganisms in organic matter has been shown to produce significant amounts of plant growth regulators including auxins (IAA), cytokinins and gibberellins (Tomati *et al.*, 1988). Similarly, Zamanov *et al.* (2002) showed that earthworm activity accelerates the process of humification of organic matter, which increases microbial populations and the accumulation of auxin- and gibberellins-like substances. Garcia Martinez *et al.* (2002) found that compost aqueous extract contained compounds with molecular structure and biological activity analogous to auxins, while Arthur *et al.* (2001) found cytokinin-like substances derived from the hydrolysis of glucosides by the enzyme β-glucosidase produced by microbes. Presence of humic, fulvic and other organic acids extracted or produced by microorganisms in VCT could induce plant growth (Arancon *et al.*, 2007).

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Assiut J. Agric. Sci., (47) No. (6-1) 2016 (87-99) *ISSN: 1110-0486*
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التأثير التثبيطى لشاى الفيرموكومبوست على العفن الأبيض فى البصل

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الملخص

تم استخدام شاى الفيرموكومبوست تحت تركيز ١، ٢، ٣، ٤، ٥ مل/أصيص (قطر ٢٥ سم) تحت ظروف الصوبة و تحت تركيز ٣، ٦، ٩، ١٢، ١٥ لتر/فدان فى توقيتات مختلفة مع ماء الرى لدراسة إمكانية مكافحة مرض العفن الأبيض فى البصل المتسبب عن فطر *Sclerotium cepivorum* Berk. تحت ظروف الصوبة والحقل بمحطة البحوث الزراعية بملوى، محافظة المنيا واستخدام مبيد الفوليكور (تيبوكونازول ٢٥%) الموصى به للمقارنة.

تحت ظروف الصوبة، أعطت معاملة شاى الفيرموكومبوست تحت تركيز ٥ مل/أصيص ثلاث أو أربع مرات مع ماء الرى أفضل أختزال لمرض العفن الأبيض على البصل بنسبة كفاءة ٥٧،١%، فى حين أن المبيد الفطرى فوليكور أعطى ١٠% أصابة مقارنةً بنسبة أصابة ٨٥،٧% لغير المعامل.

بالنسبة لتجارب الحقل، أختزلت جميع المعاملات نسبة الأصابة مقارنةً بغير المعامل. وكانت أفضل معاملة هى معاملة شاى الكومبوست بتركيز ١٥ لتر/ فدان أربعة مرات مع ماء الرى و التى أعطت نسبة أصابة ٢٠%، فى حين أعطى مبيد الفوليكور ٨،٣%. من ناحيةٍ أخرى زادت معاملة شاى الكومبوست عند تركيز ١٥ لتر/فدان مع ماء الرى محصول الأبصال كذلك أزداد العدد الكلى للبكتريا و الفطريات و الأكتينوميسيتات بالتربة تحت جميع المعاملات مقارنةً بغير المعامل.