J. Agric. Sci. Mansoura Univ., 34 (5): 5393 - 5400, 2009

COMBINED APPLICATION OF TWO *TRICHODERMA* SPP., FUSILEDE SUPER HERBICIDE AND COMPOST IN CONTROLLING *RHIZOCTONIA* DAMPING-OFF AND ROOT ROT ON SUGAR BEET

Awad H. M. F.[1]; G. A. Amer[2] and M.I. Gouda[1]

[1] **Maize, Sugar Crops and Foliages Disease Dept. Plant Pathol. Res. Instit., A. R.C., Egypt.**

[2] **Division of Plant Pathology, Faculty of Agriculture, Minufiya University, Shibin El-Kom, Egypt.**

ABSTRACT

Soil-borne plant pathogenic fungi cause heavy crop losses all over the world. Among tropical and subtropical land crops, sugar beet (*Beta vulgaris* L.) is an important crop followed sugar cane. A large number of diseases attack sugar beet plants in Egypt an all over the world .Of these, *Rhizoctonia solani*, *Fusarium oxysporum* and *Sclerotium rolfsii*. This work was carried out under field conditions of Gemmeiza Agricultural Research Station, during 2006-2007 and 2007-2008 growing seasons. In the present study, integrated management of *R. solani* of sugar beet using cominations of controlled amounts of pest-fighting micro-organisms to compost (Bio-pesticides) were effective in controlling the disease. A combined application of *Trichoderma virdi* or *Trichoderma harzianum*, Fusilede super herbicide and compost were the best. Using composted *T. virdi* + Fusilede super herbicide + *R. solani* followed by *T. harizianum* + Fusilede super herbicide + *R. solani* showed 82.05 and 81.64 % survival plants. Also, compost alone showed 59.55 survival plants comparing with *R. solani* control treatment (27.50). The results indicated that the application of these combinations successfully decrease severity and also increases the growth of sugar beet plants, sucrose and total soluble solids.

Keywords: *Rhizoctonia solani, Fusarium oxysporum , Sclerotium rolfsii, Trichoderma virdi , Trichoderma harzianum*, Fusilede super, herbicide , compost.

INTRODUCTION

Damping-off and root rot caused by *Rhizoctonia solani* and *Fusarium oxysporum*, are the most common soil-borne diseases of sugar beet .These soil born diseases are controlled by repeated applications of fungicides (seed dressing); however, these applications may result in soil contamination, fungicide resistance, or harmful effects to non-target organisms. In the United States, more than 10 percent of the vegetables planted are lost to root rot alone (according to researchers at the University of Florida's Tropical Research and Education E Center). Current fungicide options are too expensive and becoming highly regulated. Integrated management control options are required that are profitable and environmentally friendly. Compost amendments have the potential to reduce disease through building soil quality, suppressing disease organisms and improving crop root vigor. Root-rot severity varied and in some cases composted poultry manure had no significant effect but in other cases reduction in disease was observed in compost-amended soils. Integrated management root-rot organisms and soil born diseases are persistent problems that growers lack tools to control.

The effectiveness of compost and compost extracts against plant diseases caused by a wide range of pathogens and pests, including bacteria and fungi, have been demonstrated in numerous studies (Amer *et.al.*, 1997, Hoitink *et.al.*, 1993 and Ganesan *et.al.*, 2007). Various studies related to biological control of damping-off and root rots by suppressive composts have helped to diminish the incidence of these diseases around the world (Cotxarrera *et.al.*, 2002 and Kavroulakis *et.al.*, 2005). Several biocontrol agents have been identified from soils with added compost, such as *Trichoderma* (Hoitink and Boehm, 1999). The suppression capacity could be due to compost recolonization by effective biocontrol agents after peak heating occurred in the composting process (Phae *et.al.*, 1990). The objective of this experiment was to determine the plant availability of compost metals to sugar beet crop for controlling Rhizoctonia damping-off and root rot diseases.

MATERIALS AND METHODS

This experiment was carried out under field conditions of Gemmeiza Res. Sta. to study the interaction between different treatments of *Trichoderma* spp. and Fusilede super herbicide amended with compost as well as *R. solani*. A field study was conducted in which seven treatments were applied for 2 years, 2006 / 2007 and 2007 / 2008 growing seasons.

Isolation and identification.

R. solani was isolated as a mycelium from infected sugar beet roots showing symptoms of root-rot (*B. vulgaris* L.) growers in El-Gharbya Governorate. Mycelium was transferred to potato dextrose agar medium (PDA) and incubated at 28 ^{0}C. The isolated fungus was purified by using hyphal tip technique (Dhingra and Sinclair, 1995). The isolated fungus was identified by Dept. of Mycology and Pl. Diseases, Plant Pathology Inst. ARC according to Booth (1977) and Singh (1982).

Field experiment.

Inoculum of *R. solani* was prepared by the whole grain method at the rate of 0.25 % (Papavizas and De Vay, 1962). The inoculum of *R. solani* was thoroughly mixed with *Trichoderma.* spp. and compost at the rate of (1 : 1 :1) in the shared treatments.

Field treatments

Seven treatments were applied in this experiment as follow

- *T. virdi* + *R. solani.*
- *T. harizianm* + *R. solani.*
- Fusilede super herbicide + *R. solani.*
- *Compost* + *R. solani.*
- *Composted T. virdi* + Fusilede super herbicide + *R. solani.*
- *Composted T. harizianm* + Fusilede super herbicide + *R. solani.*
- *R. solani.*

Sugar beet was handily planted in hills, three rows, 3 m. length, with 0.60 m between rows and 12 hills per row (Plots measured 5.4 m.). Three seeds of Kawmera cultivar were sown in each hill which amended by compost and *Trichoderma* spp.(1:1) then covered with a thin layer of sand. The treatments were arranged in randomized complete blocks. The herbicide (Fusilede super 12.5 % EC) was used by adding to the proper quantity of water then applied with the shared treatments as spraying with enough quantity for one time. There were three control plots without compost. All treatments received fertilizer at the recommended doses.

Disease assessment

Pre-emergence damping-off was recorded after 15 days of sowing, while post-emergence damping-off was recorded after 45 days (Abd El-Moity,1986). After 150 days of planting, disease Severity (DS) was rated as percentage surface area covered by *Rhizoctonia* on sugar roots according to the scale 0 -10 adopted by (Sharma and Pathak, 1994).

Assessment of sucrose and total soluble solids.

The crop was hand-harvested after 150 days to assess average of yield (10 roots) , weight of root and root length. Sucrose % and total soluble solids (TSS%) were determined at harvest. The TSS % was determined in fresh roots using hand Refract meter (Me Ginnis, 1982). While, sucrose percentage was estimated by adding 26 gm from the minced root to 173 ml of 3 % lead acetate, shacked for 5 minutes and filtered solution was measured by Saccharo-meter as mentioned by (A. O. A. C., 1990).

Statistical Analysis and LSD values (P = 0.05) were used to detect differences among treatment means according to Snedecor *et.al.*,(1987).

RESULTS AND DISCUSSION

Data in Table (1) showed that infection was hardly affected by growing the pathogen in plots amended by *T. virdi, T. harizianum*, Fusilede super herbicide as well as compost compared with control treatment. Highly significant differences in damping- off and disease severity among all treatments. Resistance of the plants was increased by using *T. virdi* + Fusilede super herbicide + *R. solani* (82.05 survival %), followed by *T. harizianum* + Fusilede super herbicide + *R. solani* and *T. harizianum* + *R. solani* (81.64 and 76.15 survival %, respectively) then *T. virdi* + *R. solani* (75.61 survival %) and Fusilede super herbicide + *R. solani* (75.00 survival %). Compost treatment released 59.55 % survival compared with *R. solani* (27.50% survival) . The untreated control showed 96.67 % survival. Concerning with root-rot, *T. virdi* + Fusilede super herbicide + *R. solani* and *T. harizianum* + Fusilede super herbicide + *R. solani* treatments exhibited the lowest values of disease severity (2.50 and 3.33 , respectively). While, *T. virdi* + *R. solani* and *T. harizianum* + *R. solani* showed 4.00 and 4.25 disease severity comparing with Treatment of *R. solani* alone which recorded the higher value 8.33. Treatments of herbicide in the presence of *R. Solani* reduced the disease severity with average 5.00 compared with Compost amended with *R. solani* decreased disease severity to 6.00 only. Chet, (1987) stated that *T. harzianum* has been shown to act as a mycopara site against a range of economically important aerial and soil-borne plant pathogens, being successfully used in the field and greenhouse . Several investigators have examined the effect of herbicides on soil-borne pathogen and found that these herbicides decreased disease severity (Cohen; *et.al.*, 1986; Katan & Eshel, 1973; Grinstein, *et.al.*, 1984 and Al-Kherb, 1996. These results are in agreement with the results of El-Khadem and Papavizas (1984) who reported that application of linuron herbicide was found to decrease the incidence of post- emergence of cotton seedling caused by *R. solani* and *F. oxysporum* f. sp *vasinfectum* . Also, EL-Mersawy and EL-Mashad (2000) reported that all selected herbicides and hand hoeing showed clear effect on downy mildew incidence when compared with un-weeded check.

Table (1): Effect of amending compost with different combinations of *Trhicoderma* spp and Fusilede super on damping-off and root-rot of sugar beet, Gemmeiza (Average data of two experimental replications, 2006/ 2007and 2007 /2008

Treatments	Damping-off			Root rot		
	Pre-Emergence %	Post-emergence %	Surviving plants	Disease Incidence %	Disease Severity	Healthy plants
T. virdi + *R. solani*	21.36	3.03	75.61	33.66	4.00	66.34
T. harizianum + R. solani	23.85	0.00	76.15	34.64	4.25	65.36
Fusilede super + *R. solani*	28.00	0.00	75.00	45.00	5.00	55.00
T. virdi + Fusilede super + *R. solani*	14.25	3.70	82.05	26.85	2.50	73.15
T. harizianum + Fusilede super + *R. solani*	18.36	0.00	81.64	28.64	3.33	71.36
R. solani *	40.45	0.00	59.55	55.00	6.00	45.00
R. solani **	53.75	18.75	27.50	66.67	8.33	33.33
L.S.D. at 0.05 %	6.82	8.64	-	3.46	1.42	-

*Compost amended only with *R. solani* ** *R. solani* was added without compost to hills

Data in (Table 2) indicated that the root weight , root length and leaves weight of Kawmera sugar beet cultivar in all treatments were significantly affected by biocontrol agents and tested herbicide.

Highly significant differences in root weight (kg) were observed between all treatments. Sugar beet plants belonged to the treatments of composted *T. virdi* + Fusilede super herbicide + *R. solani* and composted *T. harizianum* + Fusilede super herbicide + *R. solani* showed increase by 593.20 and 305.00 % respectively. While, treatments of *T. harizianum*+ *R. solani*., *T. virdi* + *R. solani* , Fusilede super herbicide + *R. solani* and compost + *R. solani* alone were less than other treatments with the average of 184.00, 160.00. 101.20 and 50.00 %, respectively. Also, root length run in the same line. On contrast, leaves weight showed different results, since the highest values were obtained by *T. harizianum* + *R. solani* (61.61%) and *T. virdi* + Fusilede super herbicide + *R. solani* (58.09 %) increase compared with *R. solani* alone. *Trichoderma* spp. controls the pathogen but also improves the overall health of the host (Singh D., 1991 and Ganesan, 2004). A significant reduction in the incidence of root rot caused by *Rhizoctonia solani* and *Trichoderma* treatment was reported by Jayaraj and Ramabadran (1999). Chakraborty *et.al.*,(2003) reported that combined application of *Bradyrhizobium japonicum* and *Trichoderma harzianum* significantly reduced root rot disease in Soya bean.

Table (2): Effect of amending compost with different combinations of *Trhicoderma* spp. and Fusilede super on disease severity of sugar beet root-rot, root weight, shoot weight and plant height at Gemmeiza, (Average data of two experimental replications, 2006/2007and 2007 /2008.

Treatments	Disease severity	Average of root weight		root length		Shoot weight	
		kg	Increase %	cm	Increase %	kg	Increase %
T. virdi + *R. solani*	4.00	1.300	160.00	25. 00	17.20	0.833	8.74
T.harizianum + *R. solani*	4.25	1.420	184.00	25.33	18.75	1.238	61.61
Fusilede super+ *R. solani*	5.00	1.060	101.20	29.00	35.95	1.211	58.09
T. virdi + Fusilede super + *R. solani*	2.50	1.483	593.20	31.66	48.42	0.786	2.61
T. harizianum+ Fusilede super + *R. solani*	3.33	2.025	305.00	28.33	32.81	0.877	14.49
*R. solani**	6.00	0.750	50.00	23.33	9.37	0.770	0.522
*R. solani***	8.33	0.500	-	21.33	-	0.766	-
L.S.D. at 0.05%	1.42	0.18	-	2.67	-	0.13	-

*Compost amended only with *R. solani* . ** *R. solani* was added without compost to hills.

Data in Table (3) showed significant differences between values of total soluble solids (TSS), sucrose contents and yield / plot of sugar beet as affected by combined treatments of *Trichoderma* spp. and Fusilede super herbicide on controlling *R. solani.* Treatments of *T. harizianum* + Fusilede super herbicide + *R. solani* and *T. virdi* + Fusilede super herbicide + *R. solani* showed the highest values of increase % of TSS (20.46 and 15.96 %), sucrose (53.81 and 50.58%) and yield of 10 roots (305.00 and 196.60 %), respectively.

The lowest values of increase % were obtained by treatment by compost only. Now, professional growers are discovering that compost-Enriched soil can also help suppress diseases and ward off pests. Disease Control Compost technology is a valuable tool already being used to increase yields by farmers interested in sustainable agriculture. Also, beneficial uses of compost can help growers save money, reduce their use of pesticides, and conserve natural resources. Disease control with compost has been attributed to four possible mechanisms: (1) successful competition for nutrients by beneficial micro-organisms; (2) antibiotic production by beneficial micro-organisms; (3) successful predation against pathogens by beneficial micro-organisms; and (4) activation of disease-resistant genes in plants by composts. Scientists have enhanced the natural ability of compost to suppress diseases by enriching it with specific disease-fighting micro-organisms or other amendments. This amended or "tailored" compost can then be applied to crops infected by known diseases. Also, Bio-pesticides, by adding controlled amounts of pest-fighting micro-organisms to compost, are becoming effective alternatives to chemical pesticides (Kavroulakis, *et.al*, 2005).

Further research has also been undertaken to find out the mechanism of action and efficacy of the combined application of these materials at a large scale of field level.

Table(3): Effect of amending compost with different combinations of *Trhicoderma* spp. and Fusilede super on total soluble solids (TSS), sucrose contents and yield / plot of sugar beet at Gemmeiza, (Average data of two experimental replications, 2006/2007and 2007 /2008.

Treatments	Disease severity	TSS	Increase %	Sucrose	Increase %	Yield / 10 roots	
						Kg	Increase %
T. virdi + *R. solani*	4.00	16.40	11.86	14.36	40.50	13.00	160.00
T. harizianum + *R. solani*	4.25	16.13	10.02	14.05	37.47	14.20	184.00
Fusilede super + *R. solani*	5.00	16.30	11.18	15.03	47.06	10.60	112.00
T. virdi + Fusilede super + *R. solani*	2.50	17.00	15.96	15.39	50.58	14.83	196.60
T. harizianum+ Fusilede super + *R. solani*	3.33	17.66	20.46	15.72	53.81	20.25	305.00
*R. solani**	6.00	15.30	4.36	14.61	42.95	7.50	50.00
*R. solani***	8.33	14.66	-	10.22	-	5.00	-
L.S.D. at 0.05 %	1.42	0.24	-	0.36	-	3.64	-

*Compost amended only with *R. solani* . ** *R. solani* was added without compost to hills.

REFERENCES

Abd El-Moity, T.H., 1986. Further studies on the biological control of white rot disease of onion. Pd.D. Thesis, Fac. Agric. Menofiya Univ., 135 pp.

Al-Kherb, S.M. 1996. Anastomosis Groups, Pathogenicity and Sensitivity Of *Rhizoctonia solani* Isolates from Potato for Some Fungicides In The Kingdom of Saudi Arabia. Dirasat, Agriculture Sciences, 23: 208-213.

Amer, G.; Aggarwal, A. ; Singh D.V. and Srivastava, K.D., 1997. Interaction of *Bacillus thuringiensis* with *Pythium ultimum* and *Fusarium oxysporum* f. sp. *Lycopersici* : possible role in biological control. Curr. Sci. 73: 284-286.

A. O. A. C., Association of official analytical chemists 1990. Official methods analysis of the association of official analytical chemists. Washington 25. D.C., USA.

Booth, C., 1977. Fusarium laboratory guid to the identification of the major species. Commonwealth Mycological Institute, Kew Surey, Englad. 130-150.

Chakraborty, U.; Sarkar, B. and Chakraborty, B.N., 2003. Combined application of *Bradyrhizobium japonicum* and *Trichoderma harzianum* on root rot disease of soyabean. J. Mycol. Pl. Pathol., 33: 21-25.

Chet, I., 1987. In: Chet, I. (Ed.), Innovative Approaches to Plant Disease Control. Wiley, New York, pp. 137–160.

Cotxarrera, L.; Trillas-Gay, M.I.; Steinberg, C. and Alabouvette, C., 2002. Severity in okra plants, if applied prophylactically to plant suppress Fusarium wilt of tomato. Soil Biol. Biochem. 34, 467–476.

Cohen, R.; Riov, J.; Lisker, N. and Katan, J., 1986. Involvement of ethylene in herbicide induced resistance to *Fusarium oxysporum* f.sp. *melonis*. Phytopathology 76 :1281-1285.

Dhingra, O.D. and Sinclair, J.B., 1994. Basic plant pathology Methods. Second Edition, CRC press, Inc. pp 434.

El-Khadem, M. and Papavizas, G.C., 1984. Effect of the herbicides EPT and Linuron on cotton diseases caused by *Rhizoctonia solani* and *Fusarium oxysporum* f.sp. *vasinfectum*. Plant Pathol. 33 : 411-416.

EL-Mersawy, E.M. and EL-Mashad, L.A., 2000. Effect of some herbicides on weeds and downy mildew disease of maize. J. Agric. Sci. Mansoura Univ. 25 (11) : 6809-6818

Ganesan, S., 2004. Studies on the Biocontrol of Soil-borne Plant Pathogens, PhD Thesis submitted to Madurai Kamaraj University, Madurai, India.

Ganesan, S.; Ganeshkuppusamy, R. and Sekar R., 2007. Integrated Management of Stem Rot Disease (*Sclerotium rolfsii*) of Groundnut (*Arachis hypogaea* L.) Using *Rhizobium* and *Trichoderma harzianum* (ITCC - 4572) Turk J Agric For 31 (2007) 103-108.

Grainger, J., 1949. Crops and diseases. Dept. Pl.Pathol.,W. Scotland Agric. Coll. Auchineruire, Res., Bull No. 9 pp. 51.

Grinstein, A.; Lisker, N.; Katan, J. and Eshel, Y., 1984. Herbicides induced resistance to wilt diseases. Physiol. Plant Pathol. 24 : 347-356.

Hoitink, H.A.J. and Boehm, M.J., 1999. Biocontrol within the context of soil microbial communities: a substrate-dependent phenomenon. Annu. Rev. Phytopathol. 37, 427– 446.

Hoitink, H.A.J.; Boehm, M.J. and Hadar, Y., 1993. Mechanisms of suppression of soil-borne plant pathogens in compost-amended substrates. In: Hoitink, H.A.J., Keener, H.M. (Eds.), Science and Engineering of Composting: Design, Environmental, Microbiological and Utilization Aspects. Renaissance Publications, Worthington, OH, pp. 601–621.

Jayaraj, J. and Ramabadran, R., 1999. Rhizobium -Trichoderma interaction *in vivo* and *in vitro*. Indian Phytopath. 52: 190-192.

Katan, J. and Eshel, Y., 1973. Interactions between herbicides and plant pathogens. Residue Rev. 45: 147- 177.

Kavroulakis, N.; Ehaliotis, C.; Ntougias, S.; Zervakis, G.I. and Papado-poulou, K.K., 2005. Local and systemic resistance against fungal pathogens of tomato plants elicited by compost from agricultural residues. Physiol. Mol. Plant Pathol. 66, 163–174.

Me Ginnis, R.A., 1982. Beet sugar technology. 3 rd edn. Beet sugar develobment foundation for Collins, 855 pp.

Papavizas, G.C. and De Vay. C.B., 1962. Isolation and Pathogencity of *Rhizoctonia saprophytically* existing in soil. Phytopathology , 52: 834-840.

Phae, C.G.; Saski, M.; Shoda, M. and Kubota, H., 1990. Characteristic of *Bacillus subtilis* isolated from composts suppressing phytopathogenic microorganisms. Soil Sci. Plant Nutr. 36, 575–586.

Sharma, B.S. and Pathak, V.N., 1994.Yield and sucrose loss in sugar to root rot. Indian Phytopathology, 47 (4): 408-411.

Singh, B.S., 1982. Plant pathogens " The Fungi" Oxford and IBH Publishing Co. New Delhi, Bombay, Calcutta, pp.443.

Singh, D., 1991. Biocontrol of *Sclerotinia sclerotiorum* (Lib.) de Bary by *Trichoderma harzianum*. Tropic. Pest Manage. 37, 374 – 378.

Snedecor, G.W and Cochran, G.W., 1987. Statistical Methods. Oxfordl BH Publishing Co., New Delhi, pp. 20-35.

United States Solid Waste EPA 530 -F- 97- 044. Environmental Protection and Emergency Response October 1997 Agency (5306W) www.epa.gov Innovative Uses of Compost Disease Control for Plants and Animals

التأثير المشترك لعزلتين من الترايكوديرما ، مبيد الحشائش فيوزيليد سوبر والكمبوست في مقاومة فطر الريزوكتونيا المسبب لمرضي موت البادرات وعفن الجذور على بنجر السكر

حسام الدين محمد فتحي عوض[1] ، جمعه عبد العليم عامر[2] و مصطفى إبراهيم جوده[1]

[1]قسم بحوث أمراض الذرة والمحاصيل السكرية- معهد بحوث أمراض النباتات- مركز البحوث الزراعية

[2] قسم أمراض النباتات - كلية الزراعة بشبين الكوم - جامعة المنوفية

تسبب الفطريات الممرضة الكامنة في التربة خسارة كبيرة لكثير من المحاصيل في العالم من بينها محصول بنجر السكر والذي يعتبر من أهم محاصيل إنتاج السكر بعد محصول قصب السكر. يهاجم محصول بنجر السكر بكثير من تلك الفطريات الممرضة في مصر والعالم ومنها فطر الريزوكتونيا سولاني ، الفيوزاريوم أوكسيسبوريم وفطر الاسكليروشيم رولفزياي. تم إجراء هذا البحث خلال الموسمين الزراعيين ٢٠٠٦-٢٠٠٧ و ٢٠٠٧-٢٠٠٨ بمحطة البحوث الزراعية بالجميزة.

في هذه الدراسة تم استخدام خليط من الكائنات المقاومة بيولوجيا (جنس الترايكودرما) + الكمبوست كبديل للمقاومة الكيماوية حيث تم الخلط بنسب محددة (١ : ١). كما استخدم مبيد الحشائش فيوزيليد سوبر كمعاملة منفردة مع الفطر وكذلك مع المخاليط المستخدمة . أعطت معاملة بمخلوط الترايكودرما فيردي+ مبيد الحشائش فيوزيليد سوبر + الكمبوست تليها الترايكودرما هارزيانم + الحشائش فيوزيليد سوبر + الكمبوست أفضل النتائج في مقاومة المرض حيث أعطت ٨٢,٠٥ ، ٨١,٦٤ % نباتات سليمة متبقية. كذلك المعاملة بمبيد الحشائش فيوزيليد سوبر بمفرده أعطت ٥٩,٥٥ % نباتات سليمة متبقية مقارنة بمعاملة الريزوكتونيا سولاني (الكنترول) ٢٧,٥٠% نباتات سليمة متبقية .

كذلك أظهرت هذه المعاملات انخفاض شدة الإصابة بنجاح مع زيادة واضحة في نمو نباتات البنجر وكذلك زيادة في وزن وطول الجذر وزيادة وزن الأوراق. كذلك أدى استخدام هذه المعاملات إلى زيادة نسبة السكروز والمواد الصلبة الذائبة.