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SOIL SOLARIZATION: A USEFUL TOOL FOR CONTROL OF VERTICILLIUM WILT AND WEEDS IN EGGPLANT CROPS UNDER PLASTIC IN THE PO VALLEY

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SUMMARY

To develop strategies integrated with or alternative to chemicals for controlling pests, weeds and pathogens of vegetables, solarization and semi-solarization were tested in four consecutive years against *Verticillium dahliae* Kleb. on eggplants and weeds under plastic in the Po Valley, Piedmont, Italy. The temperature required for *V. dahliae* inactivation as a function of exposure time, and the number of days needed for effective solarization were also investigated. Treatment at 38-45°C for various periods were lethal to *V. dahliae* microsclerotia. The time to attain LD90 was 21, 97, 275, and 324 h respectively at 45, 42, 40, and 38°C. Temperatures and times required to kill 90% of the propagules were significantly correlated ($R^2 = -0.9437$), with a regression curve in which lethal temperature was a natural log function of exposure time. Compared with the control, semi-solarization and solarization increased the soil temperature by 8.1-10.7°C and 14.4-16.8°C respectively, at 25 cm depth. The laboratory tests suggested that in Piedmont the effective mulching period may be shorter than 45 days. Semi-solarization and solarization reduced Verticillium wilt severity by 35-48% and 89-98%, but their effectiveness was lowered by subsequent deep tillage. Eggplant yield was consistently higher in solarized than in control soil, and intermediate in semi-solarized soil. Solarization reduced weeds by 97%, whereas semi-solarization had little or only moderate herbicidal effect.

Key words: eggplant, *Verticillium dahliae*, soil disinfestation, thermal death, polyethylene.

INTRODUCTION

Production of high-quality vegetables is the main aim of modern horticulture, and development of strategies integrated with or alternative to chemicals for controlling pests, weeds and plant pathogens is an important component. The need for alternatives to chemicals for controlling soil-borne plant pathogens is underlined by the fact that methyl bromide, widely used as soil fumigant, will be dropped in 2005 because it contributes to depletion of the ozone layer (L. no. 549, 28/12/1993). Use of ecologically friendly techniques such as soil solarization to raise soil temperature to sub-pasteurisation levels, could contribute significantly to filling the methyl bromide gap.

Soil solarization was first introduced in Israel (Katan *et al.*, 1976) and later tested in other countries in hot as well temperate climates against several plant pathogens (Katan and De Vay, 1991). In Italy it has been successfully used since 1979 in mediterranean glasshouses against *Pyrenochaeta lycopersici* (Tamietti and Garibaldi, 1981, 1982) and *Rhizoctonia solani* (Tamietti and Garibaldi, 1989). In the open field solarization seems to be less effective, having been successful only against *Sclerotinia minor*, *Sclerotinia sclerotiorum*, and *Rhizoctonia solani* in Sicily and in Tuscany (Triolo *et al.*, 1985; Cartia *et al.*, 1987; Triolo *et al.*, 1989), and against several pathogens in combination with reduced dosages of Dazomet in Liguria (Minuto *et al.*, 1995).

The climate of Piedmont and the Po Valley in general is not considered fully suitable for soil solarization, as solar radiation is on average 8.5% less than in Sicily and 2.5% less than in Tuscany and Liguria (Petrarca *et al.*, 1999). However, preliminary experiments on *R. fragariae* have shown that under plastic, solarization can still be effective (Tamietti, 1994).

Eighteen percent of the total area of nearly 16,000 ha growing vegetables in Piedmont is under plastic. Solanaceous species, mainly tomato, pepper and eggplant, are normally grown in rotation with lettuce, zucchini, spinach or garden beet in such a way that the soil could easily be kept free for solarization every second year from the beginning of July to the middle of August. Summer solarization would also be possible with strawberry.

Verticillium dahliae Kleb. commonly attacking a large number of protected and unprotected crops, is

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widespread and quite damaging in Piedmont on solanaceous plants, especially the very susceptible eggplant. Control of Verticillium wilt with soil solarization has been reported in Mediterranean regions in greenhouses (Cartia, 1989; Tjamos *et al.*, 1989) and in warmer regions of the world in the open (Tjamos and Paplomatas, 1988; Morgan *et al.*, 1991; Ghini *et al.*, 1992; Melero Vara *et al.*, 1995).

Our experiments, carried out in four consecutive years on eggplants, were designed to evaluate the efficacy of solarization and semi-solarization in controlling *V. dahliae* and to determine the temperature needed for inactivation as a function of exposure time. The usefulness for weed control and the number of days required for effective solarization in the Po Valley were also tested.

MATERIALS AND METHODS

Laboratory assays. A *V. dahliae* isolate from eggplant was grown on vegetable extract agar (V8) in Petri dishes at 25°C for 6 weeks for microsclerotia production. Mycelial plugs 6 mm in diameter were then cut with a cork borer from zones rich in microsclerotia, placed in Petri dishes containing 15 ml of water agar and incubated at 38, 40, 42, and 45°C ± 0.1 for various times. Twenty disks were randomly chosen every 24 h (38 and 40°C treatments) or every 2 h (42 and 45°C), moved to fresh V8, further incubated at room temperature, and visible growth, indicating survival, recorded for 30 days. When *V. dahliae* microsclerotia are present in crop residues, quite a low inoculum density can still cause disease (Pullman and De Vay, 1982; Xiao and Subbarao, 1998). So each disk was considered as an individual and the germination scored as positive even when initiated from a single point. The experiments were done in triplicate.

For each temperature tested, the percentage of disks with viable microsclerotia (colony survival) was plotted against exposure time, and the regression curve was calculated. The temperatures tested were then plotted against the exposure time necessary to kill 90% of the propagules at each temperature as independent variable, and the LD90 regression curve was calculated and compared with the mean temperature registered in the field soil at 15 and 25 cm depth.

Field trials. Trials were carried out over five years, from 1993 to 1997, on four consecutive crops of eggplant 'Lunga violetta', susceptible to *Verticillium*, at the experiment station of the Agricultural Faculty of Turin University, Carmagnola (Turin, Piedmont), in a field selected for its history of severe Verticillium wilt. The main soil and weather characteristics are summarised in Table 1.

The soil was carefully cultivated, levelled, divided into 9 plots (6 x 5 m), and watered to saturation of the cultivated layer (25-30 cm). Three plots were covered with a transparent polyethylene film 0.10 mm thick, and 6 were left unmulched. The unmulched plots were treated with pendimethalin (0.2 ml m^{-2}) to suppress weed growth. The mulched plots (solarized) and 3 unmulched plots (semi-solarized) were protected with a plastic tunnel; 3 remaining plots not mulched and not protected were left as controls. Exposure to solar radiation was continued for at least 45 days starting July 18 in 1993, June 20 in 1994 and July 1 in 1995 and 1996. The tunnel was removed, and the soil left uncultivated until spring. To maintain a homogeneous and undisturbed inoculum potential, the site of the experiments was changed each year.

Immediately before transplanting (first week of May) the soil was fertilised ($N:P_2O_5:K_2O$ 5.7:11.5:8.4 g x m^2) and cultivated to 15-20 cm depth in 1994, 1996 and 1997, and 25-30 cm in 1995. In each plot were planted 60 'Lunga violetta' plants, previously potted in steamed soil. Soil temperature was recorded with an LSI T6 thermograph (+20 to +70°C) connected to six thermocouples inserted 15 and 25 cm deep. Yield was measured (number and weight of fruits per plant) and disease severity assessed, during growth and at harvest, on a 0-5 scale (0 = healthy plant, 1 = asymmetry and/or

Table 1. Main characteristics of soil and climate in the experiment field (1985-1995 mean data).

Soil component		Climate	June	July	August	September
Sand (%)	34	min temp. °C	13.3	16.5	15.6	11.7
Silt (%)	57	max temp. °C	25.3	28.9	28.4	23.7
Clay (%)	9	mean temp. °C	19.3	22.7	22.0	17.7
Organic matter (%)	1.53	daily insolation h/d	7.4	8.5	8.0	5.1
pH (H_2O)	7.5	solar radiation cal cm^{-2}/d	493.1	507.1	448.1	316.4
		wind km/d	58.1	42.5	38.0	34.4
		rain mm/m	67.4	38.9	42.0	64.0

chlorosis on 1-3 leaves, 2 = chlorosis and necrosis on 4-20 leaves, 3 = dwarfing, 50% defoliation, necrosis of 1-2 small branches, 4 = dwarfing, 51-80% defoliation, necrosis of small branches lower than 50%, 5 = unproductive plant, necrosis of more than 50% of the small branches.

One month after planting, the weeds were counted and determined in a 1 m^2 sub-plot per plot.

For statistical analysis, MacKinney disease indexes were calculated and converted into radians [$\theta = (\arcsin\sqrt{p}) \times 180/\pi$]. The means relative to disease severity, disease incidence, yield and weed incidence were compared by analysis of variance and Bonferroni's test.

RESULTS

Effects of heat treatments on *V. dahliae* viability in vitro. The regression curves obtained by plotting disk colony survival (%) against exposure time (h) (Fig. 1) show mortality at each temperature tested as a natural logarithm function of the time. LD90 (the exposure time at each temperature necessary to completely inactivate 90% of the disk colonies) was 21, 97, 275, and 324 h respectively at 45, 42, 40, and 38°C. The viability of the colony disks decayed after 2 ± 0.5, 62 ± 3, 120 ± 34 and 192 ± 28 h after the treatment, and continued with slopes statistically different ($P = 0.05$) only between 45 and 38°C. Temperatures and times required to kill 90% of the propagules were significantly correlated ($R^2 = -0.9437$), and gave a regression curve where lethal temperature is a natural log function of exposure times (Fig. 2).

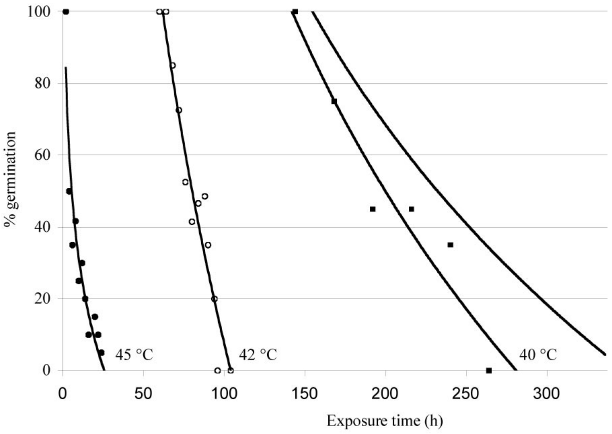

Fig. 1. Log regression of per cent survival of colony disks of *V. dahliae* on V8 agar at increasing temperature and times of exposure. Equations and statistics of regression lines for treatments at:

45°C : (107.25 ± 3.674) – (32.975 ± 8.909) Ln (x); $R^2 = 0.8995$; P value > 0.0001;

42°C : (895.95 ± 82.89) – (192.88 ± 18.99) Ln (x); $R^2 = 0.9366$; P value > 0.0001;

40°C : (826.98 ± 103.1) – (146.70 ± 19.45) Ln (x); $R^2 = 0.9343$; P value > 0.0017;

38°C : (721.92 ± 54.65) – (123.35 ± 10.02) Ln (x); $R^2 = 0.9558$; P value > 0.0001.

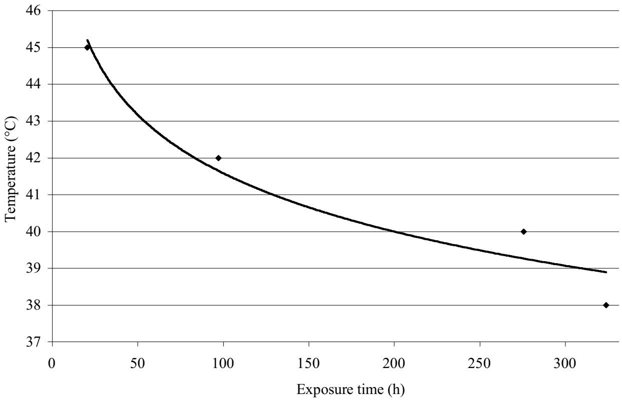

Fig. 2. LD90 curvilinear regression relative to *V. dahliae* in V8 agar: temperature of exposure required for 90% propagule inactivation as a function of time.

Field trials. At 25 cm depth, the mean soil temperature in the control plots was 25-26°C in 1993, 1995, 1996 and 27-28°C in 1994. Semi-solarization and the solarization raised this temperature by 8.1-10.7°C and 14.4-16.8°C respectively (Figs 3 and 5). In the solarized soil the temperature exceeded 40°C for about 780 h in 1995 and 1996 and 964 h in 1997 at 15 cm depth; 797 h in 1996, and 868 h in 1994, at 25 cm depth. Down to 15 cm the soil temperature exceeded 50°C for about 40 h in 1994 and 1996. In the semi-solarized soil the temperature was higher than 40°C for 258 h in 1994, 112 h in 1995 and 71 h in 1996, only at 15 cm depth. Further data are reported in Table 1.

In the bare soil the incidence of Verticillium wilt was very high (more than 99.0%) in 1994 and 1995, moderate in 1997 (65.0%) and low in 1996 (14.7%) and the severity ranged 45.6-56.5%, with the exception of 1996, when it was 7.2%. In the semi-solarized soil, disease incidence and severity were significantly lower ($P = 0.01$ in 1994 and 1997; $P = 0.05$ in 1995) than in the control in three years out of four. In the solarized soil 2-8% of the plants were infected with a severity ranging from 1 to 2% in three trial out of four (Fig. 6). In 1995 disease incidence was 51% and disease severity 18.3% (Table 2).

Yield was consistently higher in the solarized plots (8.9-11.5 berries per plant, and total weight of 3.7-4.0 kg), intermediate in the semi-solarized soil, (7.5-9.8 fruits, 2.6-3.2 kg), and lower in the control soil (7.5 fruits, 2.0 kg) (Table 3).

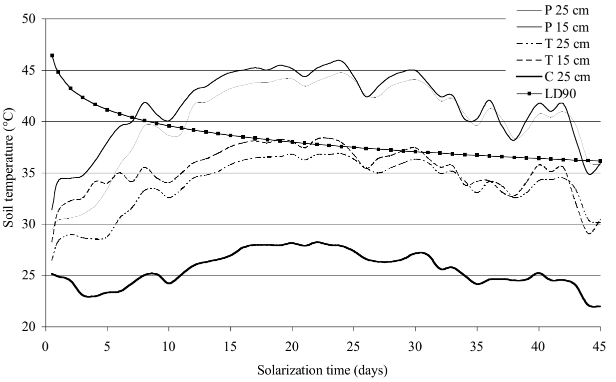

Fig. 3. Mean temperature in the solarized (s), semi-solarized (s-s) and control soil at 15 and 25 cm depth in 1995, and theoretical LD90 of *V. dahliae* in response to heat treatments.

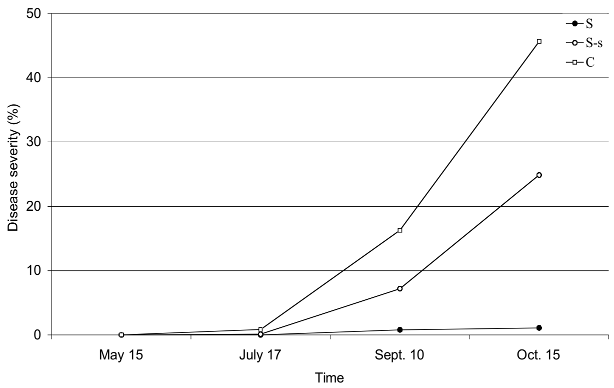

Fig. 6. Verticillium wilt severity progression in eggplants grown in solarized (s), semisolarized (s-s) and control (c) plots, in 1997.

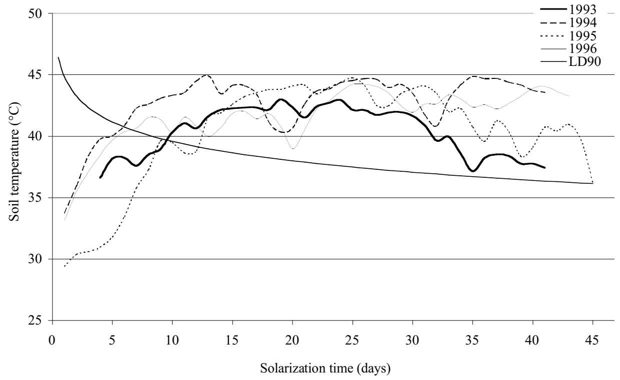

Fig. 4. Mean temperature in the solarized soil at 25 cm depth during the 4 years of experiments (1993-1996), and theoretical LD90 of *V. dahliae* in response to heat treatments.

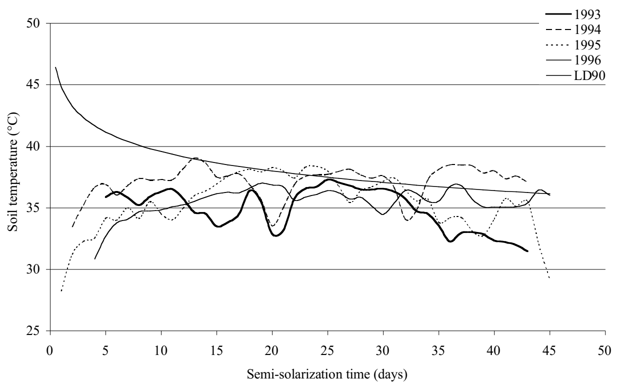

Fig. 5. Mean temperature in the semi-solarized soil at 15 cm depth during the 4 years of experiments (1993-1996), and theoretical LD90 of *V. dahliae* in response to heat treatments.

Table 2. Hours of exposure of the soil to different temperatures at 15 and 25 cm depth during the solarization and semi-solarization treatments.

Year	Treatment/ depth (cm)	Hours with T > °C				
		38	40	42	45	50
1993	semi-sol/25	12	0	0	0	0
7/18-8/31	solariz/25	830	680	458	5	0
1994	semi-sol/15	415	258	88	0	0
6/20-7/25	semi-sol/25	152	0	0	0	0
	solariz/15	992	964	821	403	40
	solariz/25	952	868	694	238	0
1995	semi-sol/15	280	112	10	0	0
7/1-8/16	semi-sol/25	18	0	0	0	0
	solariz/15	938	780	576	260	0
	solariz/25	924	724	492	100	0
1996	semi-sol/15	293	71	0	0	0
	semi-sol/25	64	0	0	0	0
7/1-8/16	solariz/15	947	780	569	339	41
	solariz/25	943	797	497	82	0

In control plots the weeds numbered 236 to 615 plants m^{-2}, with average composition of *Portulaca oleracea* (40.0%), *Galinsoga parviflora* (21.6%), *Chenopodium polyspermum* (21.1%), *Echinochloa crusgalli*, *Solanum nigrum*, *Stellaria media*, *Amaranthus retroflexus* (1-3% each), with 6.2% of 20 other species changing each year in presence and frequency.

Semi-solarization significantly ($P = 0.05$) reduced the population of *Chenopodium polyspermum*, *Galinsoga parviflora*, *Stellaria media* and *Solanum nigrum*, in two trials out of four. Solarization was effective against all weed species, reducing their number by 66% in 1994

Table 3. Incidence and severity (%) of Verticillium wilt of eggplant at the end of the growing season in solarized, semi-solarized and bare (control) soil during the 4 years of experiments.

Survey date	Treatment					
	Solarization		Semi-solarization		Control	
	Incidence	Severity	Incidence	Severity	Incidence	Severity
19/9/1994	8.0 c*	2.0 c	72.2 b	25.6 b	100 a	49.3 a
21/9/1995	51.0 c	18.3 c	83.0 b	31.6 b	99.1 a	56.5 a
26/9/1996	1.9 b	0.8 b	11.5 ab	4.7 ab	17.4 a	7.2 a
15/10/1997	2.4 c	1.1 c	44.2 b	24.8 b	64.5 a	45.6 a

* Means on the same row relative to disease incidence or severity followed by the same letter are not statistically different (small letters $P = 5\%$; capital letters $P = 1\%$) following Bonferroni's test.

Table 4. Yield (berry number and weight per plant) for eggplants grown in solarized, semi-solarized and bare soil during 4 years of experiments.

Year	Treatment					
	Solarization		Semi-solarization		Control	
	no.	Kg	no.	Kg	no.	Kg
1994	10.6 a*	4.04 a	7.5 b	2.63 b	6.0 c	1.97 c
1995	8.9 a	3.60 a	8.4 b	3.19 b	6.0 c	1.80 c
1996	5.3 A	1.82 a	4.2 b	1.46 b	4.6 ab	1.68 ab
1997	11.5 A	3.69a	9.8 ab	3.15 a	7.5 b	2.07 b

* See Table 3.

and by 97% in the other years. Solarization did not eradicate *Portulaca oleracea* and *Digitaria sanguinalis*; *Taraxacum officinale* and *Solanum nigrum* also persisted to some extent (0.3-1 plant m^{-2}) in three experiments out of four (Table 4).

DISCUSSION

The thermal-death curves drawn by plotting the responses of *V. dahliae* to heat treatments were log functions of the exposure time, as previously shown for *V. dahliae* and other fungi (Munnecke *et al.*, 1976; Pullman *et al.*, 1981; Nicolotti *et al.*, 1998), and predict the exposure time necessary to kill the fungus at constant temperatures. On the basis of the LD90 obtained with the laboratory tests, propagules of the pathogen were subjected in our solarized plots to heat dosages far exceeding those required for inactivation. This conclusion is strengthened by the fact that *V. dahliae* microsclerotia on artificial media are more resistant to heat than those produced in natural soil (Pullmann *et al.*, 1981). Thus, in accordance with results obtained on other pathogens in Liguria by Minuto *et al.* (1995), the mulching period could also be shortened in Piedmont.

In the climatic conditions of the Piedmont sector of the Po Valley, as in warmer locations (Grinstein *et al.*, 1979; Hardy and Sivasithamparam, 1985; Tjamos and Paplomatas, 1988), full solarization markedly decreased the incidence of Verticillium wilt in three out of four years. Its lower efficacy in 1995 was probably due to the fact that, unlike the other years, the soil was rototilled to 30 cm depth after the treatment, probably raising living inoculum to the soil layers explored by the plant roots. Thus, to optimise the effectiveness of soil solarization in Piedmont, the soil must not be disturbed after treatment.

In the semi-solarized plots (simple closure of the plastic-house), although the time-temperature dosages were always lower than those necessary to kill the fungus *in vitro*, the treatment was partially effective. This was probably due to inactivation of inoculum in the upper soil layers where the temperature approached 40°C and to cumulative damage to the fungal propagules, gradually affecting individual cells in the microsclerotia (Pullman *et al.*, 1981), making them more sensitive to biological control agents (De Vay and Katan, 1991). Indeed, germination of microsclerotia exposed to sublethal heat dosages was delayed, as reported in a previous work (Pullman *et al.*, 1981).

In treated soil, yield improved in quantity and quality proportionally to the plant health. No effect was noticed on the timing of fruit ripening. The poor yield obtained in 1996 in all experiments was due to a heavy

Table 5. Weed infestation (number of plant m^{-2}) observed 45 days after eggplant plantation in solarized (S), semi-solarized (S-S) and untreated control soil (C).

Species	1994			1995			1996		1997		
	S	S-S	C	S	S-S	C	S	S-S	S	S-S	C
Amaranthus retroflexus	4.3	21.0	13.5		5.0	10.9	0.2	14.7		2.3	3.3
Capsella bursa-pastoris					7.4	25.1				3.7	0.7
Chenopodium album	0.7	83.3	7.0		19.0	32.0		28.3	2.0	19.7	6.7
Ch. polyspermum	8.0	43.7	131.0	0.8	13.7	158.7				18.0	2.0
Cirsium arvense	0.3					0.5					
Digitaria sanguinalis	1.0	22.7	2.0	0.5	0.5	4.2	0.2	2.0	0.3	2.7	2.7
Diplotaxis muralis	0.7	3.3	3.0								
Echinochloa crus-galli		4.3	2.5		3.7	4.8	0.2	5.0	0.3	2.0	10.0
Equisetum arvense								0.5			
Erigeron canadensis								0.2			
Galinsoga parviflora		101.7	82.0	0.8	17.5	76.5		1.8	1.3	26.7	139.7
Lamium purpureum					0.3	1.9		0.2		0.7	
Linaria sp.		1.3	8.0								
Lithospermum arvensis		5.3									
Medicago lupulina					0.3						
Oxalis sp		1.0	16.0		0.5	2.4					
Panicum dichotomiflorum	0.3		0.5				0.2	0.5			
Papaver rhoeas		0.3									
Plantago major	0.3	11.0	4.5			7.9		0.2			
Poa annua		4.3		0.8	38.4	8.7				7.7	
Polygonum aviculare		5.3									
Polygonum persicaria	85.7	23.0	6.5			0.5		7.0			
Portulaca oleracea	82.0	192.7	209.5	12.2	104.7	240.2	3.4	23.0	1.3	131.7	61.7
Solanum nigrum	0.7	4.3	16.5	0.3	1.8	14.8	0.2	2.2		2.3	
Sonchus oleraceus						1.3		0.2		0.3	
Sorghum halepense						1.3		3.7			
Stellaria media		6.3	14.0		0.5	12.2				0.7	9.3
Taraxacum officinale	1.0	1.3	1.0	1.3	1.3	4.5			0.3	0.3	0.3
Trifolium repens		0.3	0.5								
Urtica dioica					0.3						
Veronica persica		2.0	4.0		0.3	0.8					
Asteraceae						0.5					
Cruciferae								0.2		0.7	
Labiatae					3.7	5.3					
Not det.	0.7	6.0	9.0				1.9	4.5	0.3	0.7	
Total	185.7	544.6	531.0	16.6	219.0	615.1	6.3	94.0	6.0	220.1	236.4
$P = 0.01$	A	B	B	A	B	C	A	B	A	B	B

hail-storm in July. The hail probably also affected disease expression, because symptoms were late and slight even in the control.

Soil solarization appeared to be a good method for controlling weeds. It gave excellent results, reducing the weed population by about 97% in three years out of four; in 1994 it partially failed, reducing the number of weeds by only 65%, as a consequence of the lower soil temperature obtained in 1993. On the contrary, in spite of previous treatment of unmulchened plots with pendimethalin, the herbicidal effect of semi-solarization was weak, even if sometimes statistically significant.

Several weeds, *P. oleracea* in particular, but also *D. sanguinalis*, *T. officinalis* and *S. nigrum* appeared to be quite resistant to solarization. Conflicting results have been reported on the susceptibility of *P. oleracea* and *D. sanguinalis* to solar heating (Elmore, 1991). However it is quite difficult to compare the results obtained by different workers, because many factors could affect the sensitivity of the seeds, such as soil moisture, temperature level and fluctuation, type of plastic film, tarping duration, and seed distribution in the soil profile. In practical terms, chemical treatment was not necessary, following solarization.

The temperatures registered in the treated soil were similar to those previously observed in the same location (Tamietti, 1994), suggesting that in Piedmont this method of soil disinfestation can be successful against Verticillium wilt during all the summer months. However its early application in summer is recommended in order to exploit its potential herbicidal effect.

We conclude that soil solarization is effective against Verticillium wilt and weeds under plastic in northern Italy. As it carries low ecological impact, it should be regarded as an alternative to traditional soil disinfestation methods. Where soil mulching is not considered economic, the farmers should be advised to keep their plastic tunnels closed during the summer after an herbicidal treatment of the soil, in order to profit from the partial benefits of a semi-solarization.

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