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R. Paul Schreiner · Kelly L. Ivors · John N. Pinkerton

Soil solarization reduces arbuscular mycorrhizal fungi as a consequence of weed suppression

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Abstract Soil solarization, the process of heating soil by covering fields with clear plastic, is a promising method to reduce populations of soilborne pests and weeds without the use of pesticides. However, the destruction of beneficial organisms such as arbuscular mycorrhizal (AM) fungi also may occur, thereby reducing positive effects of solarization. We compared the effects of solarization and chemical fumigants on the survival of indigenous AM fungi in 1995 and 1996. The infectivity of AM fungi was monitored before and after solarization using a greenhouse bioassay with *Sorghum bicolor* L. for both years. AM colonization of roots was also monitored in the field 8 months after solarization in 1995. Weed densities were measured 8 months after treatment in 1996. Solarization increased the average daily soil temperature 6–10°C and the maximum soil temperature reached by 10–16°C (5–20 cm depth). Solarization did not reduce the infectivity of AM fungi immediately after the solarization period in either year, as determined by the greenhouse bioassay. Infectivity was greatly reduced in solarized plots 8 months after solarization (over winter) in both years as assessed in the field (1995) or with the greenhouse bioassay (1996). Fumigation with metam sodium at 930 l ha^{-1} (350 kg active ingredient ha^{-1}) reduced the infectivity of AM fungi in both years, and fumigation with methyl bromide at 800 kg ha^{-1} eliminated infection by AM fungi. Solarization was as effective as methyl bromide and metam sodium at 930 l ha^{-1} in controlling winter annual weeds measured 8 months after treatment. Solarization apparently reduced AM fungi in soil indirectly by reducing weed populations that maintained infective propagules over the winter. Fumigation with metam sodium or methyl bromide directly reduced AM fungi in soil.

R.P. Schreiner (✉) · K.L. Ivors · J.N. Pinkerton
USDA, Agricultural Research Service,
Horticultural Crops Research Laboratory,
3420 NW Orchard Ave., Corvallis, OR 97330, USA
e-mail: schreinr@bcc.orst.edu
Fax: +1-541-7508764

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Introduction

Balancing the need to control soilborne pathogens and weeds in agricultural and horticultural systems against the environmental hazards of pesticide use is an immense challenge for researchers and farmers alike. Although chemical control of many soilborne pathogens is both effective and economical, alternative control measures to replace or reduce pesticides are sought. Methyl bromide is effective in controlling many plant pathogens and weeds, but its use will be banned as a consequence of its role in ozone depletion. Metam sodium (which breaks down to methylisothiocyanate in soil) is a common replacement for methyl bromide. Solarization is the process of heating soil by capturing the sun's energy under clear plastic. The resulting increase in soil temperatures can reduce the populations of soilborne plant pathogens and weeds (Katan and DeVay 1991). Solarization may be a useful environmentally benign alternative to chemical fumigants

Soil fumigants are known to suppress AM fungi under most circumstances (Trappe et al. 1984). Metam sodium at 40 ppm, equivalent to 140 kg active ingredient (a.i.) ha^{-1}, eliminated mycorrhiza formation when applied to sterile soil with AM fungus spores added (Nesheim and Linn 1969); however, metam sodium had little or no effect on arbuscular mycorrhizae when applied to a nursery soil at the rate of 280 kg a.i. ha^{-1} (Linderman 1987). Fumigants releasing methylisothiocyanate in soil alone (Dazomet) or in combination with other chemicals (Vorlex) have given mixed results on mycorrhizae when applied at rates of 200–1,000 kg ha^{-1} (Mark and Cassels 1999; Ocampo and Hayman 1980; Riffle 1980; Robertson et al. 1988).

Solarization can control many soilborne plant pathogens and reduction in pathogen populations and corresponding crop yield increases may last over a number of

growing seasons (Katan and DeVay 1991; Pullman et al. 1981). The focus of early solarization work was to control *Verticillium dahlia* L., which causes wilt diseases of many crops (Katan et al. 1976). Other soilborne pathogens controlled by solarization have included fungal pathogens such as *Pythium*, *Fusarium*, *Sclerotium*, and *Rhizoctonia* species, plant parasitic nematodes such as *Mesocriconemella*, *Meloidogyne*, and *Pratylenchus* species, and the bacterial pathogen *Agrobacterium tumefaciens* (Stapleton and DeVay 1986). An added benefit of controlling plant pathogens by solarization is that weeds are suppressed, particularly winter annuals (Elmore 1991; Peachey et al. 2001).

Less well understood is the effect of solarization on beneficial organisms, including mycorrhizal fungi. Bendavid-Val et al. (1997) found a significant decrease in AM fungus propagules and in the mycorrhizal colonization of onion and carrot roots growing in solarized soils. Decreased plant growth was associated with reduced mycorrhizal colonization of these crops. Pullman et al. (1981) also found a decrease in AM fungus propagules after solarizing soil at one site, but not at another. The reduction of AM fungi was associated with higher soil temperatures attained in solarized plots at the first site compared with the second. Other studies have shown either no effects or increased AM colonization in solarized soils that reached similar temperatures to those at which suppression of AM fungi was found (Afek et al. 1991; Nair et al. 1990). Solarization essentially eliminated ectomycorrhizal fungi from a forest nursery within the upper 15 cm of the soil profile (Soulas et al. 1997).

Different effects of solarization on mycorrhizal fungi may be related to different tolerances of heat stress among AM fungus species. Different temperature optima for germination and colonization of roots are known for different AM fungi (Haugen and Smith 1992; Tommerup 1983b). Temperatures lethal to AM fungi also may vary between species or isolates (Bendavid-Val et al. 1997; Menge et al. 1979; Vogelzang et al. 1993). Another possible explanation for the differences found in response to solarization is that spores are more resistant to heat stress than hyphal fragments, such that a preponderance of hyphal fragments instead of spores could increase susceptibility to heat stress (Bendavid-Val et al. 1997).

The purpose of this present study was to compare the effects of solarization and fumigation with metam sodium or methyl bromide on the infectivity of indigenous AM fungi under field conditions. The impact of our treatments on selected plant pathogens and on annual bluegrass is presented in Pinkerton et al. (2000) and Peachy et al. (2001), respectively.

Materials and methods

The potential use of solarization to control a number of plant pathogens under the climatic conditions of Oregon was examined. Field plots (2×2 m) were set up at the Oregon State University, Botany Research Farm in 1995 and 1996 on a fine-silty, mixed, mesic, Cumulic Ultic Haploxeroll. Land use prior to our study was a weedy fallow for 4 years. During this time weed populations were controlled by cultivation 3–4 times per year, which resulted in minimal plant cover during the dry season (June–October) and extensive weed cover during the winter and spring. In 1995, the impact of solarization and two rates of metam sodium fumigation (four treatments) on the infectivity of AM fungi were examined. In 1996, we assessed fumigation with methyl bromide in addition to the four treatments examined in 1995. Each treatment plot (2×2 m) was replicated four times in 1995 and five times in 1996 in a randomized complete block design. Root colonization data were arcsine transformed, analyzed by ANOVA, and mean contrasts were made using Fisher's protected LSD at 95% confidence.

Field plots and treatments

Plots were tilled in the spring of each year and fallowed until midsummer. One day prior to treatment, the field was irrigated with 2.5 cm of water via overhead sprinklers. Metam sodium (Vapam, ICI Americas, Wilmington, Del.) was applied at 230 or 930 l ha^{-1} (equivalent to 90 or 350 kg a.i. ha^{-1}) and the soil was rotovated and surface sealed by rolling. All other treatment plots were rotovated at this time. Solarized plots were overlayed with clear plastic (0.6 mil transparent polyethylene) held in place by burying the edges. Control plots received no further treatment. The solarization period lasted from 21 July to 19 September (59 days) in 1995 and from 26 July to 18 September (53 days) in 1996. Soil temperatures were monitored hourly at 5, 10 and 20 cm depths in one randomly selected solarized and non-solarized plot. Thermistors were placed at the indicated depths in the center of each plot and temperatures were recorded hourly with a CR21 micrologger (Campbell Scientific, Logan, Utah). Control and metam sodium-treated plots were kept weed free by hand weeding during the solarization period in both years. Methyl bromide (Great Lakes Chemical Corp., West Lafayette, Ind.) was applied at the rate of 800 kg a.i. ha^{-1} under a plastic tarp in 1996 only. The tarp was removed after 5 days.

Sampling

Soil samples were collected from 0–30 cm depth by combining eight randomly selected cores (2.5 cm diameter) per replicate plot. Samples were collected immediately before and after the solarization period (20 July and 19 September, 1995 and 25 July and 18 September 1996) and again 8 months after treatment. Soil samples were thoroughly mixed and stored moist at 4°C for up to 30 days. A 200 g subsample was removed from the bags, air dried at room temperature, and stored dry at 4°C for another 28–32 days to overcome possible dormancy of spores or other propagules of AM fungi (Tommerup 1983a).

Infectivity assays for AM fungi

A greenhouse bioassay was used to estimate the infectivity of AM fungi before and after the solarization period for both years and 8 months after treatment in the 1996 trial. Air-dried soil samples from each replicate plot were passed through a 2-mm sieve, uniformly mixed, diluted with nine parts autoclaved soil, and placed in duplicate 150 ml Conetainers (Stuewe, Corvallis, Ore.). Five-day-old sorghum seedlings were transplanted into the containers and grown in the greenhouse for 21 days with supplemental light provided by 1,000-W metal-halide lamps (600 μE m^{-2} s^{-1} PAR added by lamps) on a 16-h photoperiod. After 21 days, the entire root system of each plant was washed free from soil, and roots were cleared and stained for mycorrhizal colonization using standard methods (Schreiner and Bethlenfalvay 1997). The percentage of the root length colonized by AM fungi (based on the presence of arbuscules, vesicles, or hyphae connected to arbuscules or vesicles) was determined by the grid line intercept method (Newman 1966) under a dissecting microscope.

A field assay was used to assess the impact of treatments on mycorrhiza formation 8 months after the solarization period in the 1995 trial. The field assay was conducted during the late spring of 1996. On 10 May, four 7-day-old sorghum seedlings were transplanted into each of the field plots and grown for 56 days. Irrigation was applied as needed via overhead sprinklers. Plants were subsequently dug from the plots to a depth of 30 cm and fine roots were randomly sampled from each root system. Colonization by mycorrhizal fungi was determined as above.

Weed densities were recorded in the 1996 trial 8 months after treatment during the spring of 1997. On 12 May, a 0.25-m^2 grid was randomly placed in each treatment plot and individual plants within the grid were counted and identified to genus.

Results

The effects of solarization on soil temperatures at three depths are shown in Table 1. In 1995, the average temperature of soil 5–20 cm deep was 8–10°C higher in solarized plots than in non-solarized controls. In 1996, the average temperature was 6–8°C higher in solarized than in control plots. The maximum soil temperature from 5–20 cm depth was 10–15°C higher in 1995 and 11–16°C higher in 1996 in solarized than in control plots. An estimate of the thermal dose that occurred in solarized plots is shown as the cumulative hours of temperatures above 35, 40 and 45°C (Table 1). Solarization effects on soil temperatures were similar over the 2 years of our study.

Treatment effects on AM fungi were initially examined (before and after solarization in 1995) using the infection unit method of Franson and Bethlenfalvay (1989), but treatment effects were identical to the percentage root colonization at 1:10 dilution and this was used thereafter (data not shown). The effects of solarization and fumigation with metam sodium on the infectivity of AM fungi were consistent for both years of our study (Tables 2 and 3). Solarization did not significantly affect the infectivity of AM fungi in soil immediately after the solarization period in either year. The infectivity of AM fungi was dramatically reduced in both years 8 months after solarization (over winter). In 1995, we examined field-grown plants 8 months after treatment and found

Table 1 Soil temperatures (mean and maximum, °C) recorded hourly at three depths (cm) in solarized and non-solarized field plots during the solarization periods in 1995 and 1996

Solarization period	Depth	Soil temperature (°C)				Cumulative hours in solarized plots		
		Non-solarized		Solarized		>35°C	>40°C	>45°C
		Mean	Max	Mean	Max			
21 July–19 Sept. 1995	5	23.2	34.5	31.7	49.3	399	195	53
	10	23.4	31.2	33.1	45.9	445	160	9
	20	22.8	27.4	31.0	37.4	75	0	0
26 July–18 Sept. 1996	5	22.9	37.0	29.3	52.6	309	169	66
	10	23.6	32.7	31.7	47.6	390	152	17
	20	22.4	28.0	29.9	39.1	101	0	0

Table 2 Effects of solarization and fumigation in 1995 on arbuscular mycorrhiza (AM) colonization (%) of *Sorghum bicolor*. Values represent means (SE). Means within a column followed by the same letter are not significantly different ($P \leq 0.05$)

Treatment	Colonization		
	Greenhouse assay (n=8)		Field assay (n=16) June 1996
	July 1995	September 1995	
Control, non-solarized	2.48 (0.63)	2.73 (0.88) a	12.9 (1.16) a
Solarized		2.25 (0.59) a	2.8 (0.80) b
Metam sodium (low)		1.17 (0.34) ab	8.8 (1.20) ab
Metam sodium (high)		0.51 (0.18) b	6.9 (1.83) b
ANOVA (P)		0.032	<0.001

Table 3 Effects of solarization and fumigation in 1996 on AM colonization (%) of *Sorghum bicolor* in the greenhouse assay and on weed densities (number per $m^{2)}$ in field plots. Values represent means (SE). Means within a column followed by the same letter are not significantly different ($P \leq 0.05$)

Treatment	Colonization (n=10)			Weed density May 1997
	July 1996	September 1996	May 1997	
Control, non-solarized	1.69 (0.37)	1.97 (0.61) a	3.66 (0.65) a	357 (48) a
Solarized		1.74 (0.36) a	0.10 (0.06) b	36 (7) b
Metam sodium (low)		0.83 (0.43) b	0.78 (0.38) b	352 (75) a
Metam sodium (high)		0.08 (0.09) b	0.06 (0.08) b	107 (20) b
Methyl bromide		0 (0)	0 (0)	38 (9) b
ANOVA (P)		0.004	<0.001	<0.001

that solarized plots had the lowest levels of root colonization by AM fungi, control plots the highest, and metam sodium plots were intermediate (Table 2). This was surprising because soil from metam sodium plots had the lowest infectivity of AM fungi immediately after solarization. In 1996, using the greenhouse assay at all sampling times, we found the infectivity of AM fungi also was reduced 8 months after solarization, even though no effect was apparent immediately after solarization (Table 3).

A distinctive checkerboard pattern could be seen in our experimental plots in the spring following treatment because of the dramatic effect on weed populations. In the 1996 trial, weed densities measured in plots 8 months after solarization showed the strong suppression of weeds that occurred in solarized soil over the winter (Table 3). Weeds were almost eliminated in solarized plots and those treated with methyl bromide. Plots treated with metam sodium at the low rate had similar weed densities as the control. No significant differences in the diversity of weeds present in different treatments were found (data not shown). The dominant weeds present in our plots were annual bluegrass (*Poa annua* L.), Canadian thistle (*Cirsium arvense* L.), false dandelion (*Pyrrhopappus* sp.), pineapple weed (*Matricaria matricaroides* L.), wild geranium (*Geranium oreganum* Howell), foxtail (*Setaria* sp.), speedwell (*Veronica* sp.), and shephard's purse (*Capsella bursa-pastoris* L.). Mycorrhizal host plants comprised 86–95% of all weeds growing in plots 8 months after treatment, with only shephard's purse lacking mycorrhizae.

Fumigation of soil with metam sodium at the high rate significantly reduced the infectivity of AM fungi in both years when measured immediately after the solarization period and the following spring (Tables 2 and 3). Metam sodium applied at the low rate reduced the infectivity of AM fungi in 1996, but not in 1995. Differences between the low and high rates of metam sodium were not statistically significant in either year. Methyl bromide completely eliminated AM fungus propagules to our sampling depth of 30 cm. (Table 3).

Discussion

Solarization increased soil temperatures in our study to levels similar to those achieved in warmer climates (Katan and DeVay 1991). The temperatures reached by solarizing soil in the midsummer in Oregon were 10–15°C higher than control plots and resulted in significant reductions of important plant pathogens (see Pinkerton et al. 2000). Solarization did not reduce AM fungi in our soil just after solarization, although similar soil temperatures resulting from solarization have reduced AM fungi in other studies (Bendavid-Val et al. 1997; Pullman et al. 1981).

Of interest in our study was the large decline in the infectivity of AM fungi that occurred in solarized plots over the winter. We suspected that this decline was due to suppression of weeds in solarized plots. Weed populations in solarized plots were reduced to roughly one-tenth of the control level in 1996. Thus, it seems that solarization may have indirectly reduced AM fungus propagules by suppressing weeds that would maintain AM fungi over the winter. The importance of winter plant cover to maintain AM fungi in soil has been demonstrated by the positive effects that winter cover crops have on AM fungus spores and root colonization of subsequent, main season crops (Boswell et al. 1998; Galvez et al. 1995). In our case, because no cover crops were included, the maintenance of AM fungi depended on weeds. This is supported by two other observations from our study. First, the propagule densities of AM fungi at this site were relatively low, as shown by the low levels of colonization in our infectivity assays, suggesting that any plant cover would be of benefit in boosting or maintaining mycorrhizal propagules. Secondly, in 1996 when the greenhouse assay was used for all sampling dates, the infectivity of AM fungi increased over the winter in control plots (when weeds were present), but did not increase over the summer (when plots were kept weed free). Weeds that had grown over the winter in plots fumigated with the low rate of metam sodium did not boost the infectivity of AM fungi because the propagule density was already too low to be enhanced by the weed cover.

Another possible explanation of our results is that sublethal heat stress weakened AM fungus propagules in solarized plots and reduced their survival over the winter. We cannot rule out this mechanism, but we believe that sublethal heat stress would have been expressed in our bioassay immediately after solarization because the soil samples collected for the AM bioassay were stored for 4 weeks at 4°C, then air-dried and stored for another 4 weeks at 4°C. The reported effects of sublethal heat stress on other soilborne fungi occurred over relatively short periods of 1–3 days (Freeman and Katan 1988; Lifshitz et al. 1983). Additionally, populations of the fungal pathogens *Verticillium dahlia* and *Phytophthora fragaria* did not show further decline in our solarized plots over the winter compared with just after solarization (see Pinkerton et al. 2000).

The lack of a solarization effect on the infectivity of AM fungi just after solarization may indicate that most AM fungus propagules were present as spores or colonized root fragments and not hyphal fragments, which are more sensitive to heat stress (see Bendavid-Val et al. 1997). Another possibility is that the species of AM fungi present in our soil were more tolerant of high soil temperatures. Our findings do not reveal why solarization has variable effects on mycorrhizae, but show that weed suppression can be a factor influencing the effect of solarization on AM fungi. The importance of weeds to maintain AM fungi found in our study may not occur in other soil systems where AM fungus propagules are very abundant.

The results with metam sodium, where even a low rate of metam sodium reduced AM fungi under field conditions, support work by Nesheim and Linn (1969). Our data indicate that metam sodium is not a good alter-

native to methyl bromide for reducing the negative impact on mycorrhizal fungi, as suggested by Linderman (1987). The high rate of metam sodium appeared to be necessary for pathogen (Pinkerton et al. 2000) and weed control (Peachy et al. 2001) in our study. Metam sodium applied at the high rate was not significantly different from methyl bromide in reducing AM fungi. The apparent elimination of AM fungi in methyl bromide treated soil is well known (Bendavid-Val et al. 1997; Jawson et al. 1993; Menge 1982).

Solarizing soil during the summer may not be economical for vegetable growers in the Willamette Valley of Oregon, because of the loss of income during the growing season. Solarization during the summer is feasible for a number of horticultural and nursery crops, however, because fall or spring planting is often preceded by a summer fallow period.

The results of our study show that soil solarization per se does not reduce propagules of AM fungi in our soil, but rather it reduces weeds that maintain AM fungi over the long-term. Solarization may not impede mycorrhiza formation as long as crops are quickly planted into solarized soils. We would not recommend solarization followed by a long fallow period prior to planting crops dependent on mycorrhizal fungi.

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