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Steam and exothermic reactions as alternative techniques to control soil-borne diseases in basil

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Abstract – The search for non-chemical methods to control soil-borne pathogens has recently intensified ahead of the forthcoming phase-out of the fumigant methyl bromide in 2005. Indeed, none of the previously available alternatives match methyl bromide wide-spectrum control. Here, we tested two pre-planting treatments: steaming with a soil-steaming machine and exothermic reaction by incorporation into the soil of potassium hydroxide (KOH), a substance that causes an exothermic reaction with water. Experiments were conducted from 2003 to 2005 in open-field conditions by assessing the effectiveness of steam and exothermic reaction against *Fusarium oxysporum* f.sp. *basilici* and *Sclerotinia minor* on basil. The results show that steam combined with exothermic reaction reduce the incidence of *Fusarium* wilt (77–96%) better than steam only (70–89%). The control of *Sclerotinia minor* was better with potassium hydroxide (94–92%) than with steam only (83–87%). Further, we evaluated the effects of the treatments on microbial population, yield and weed control. Here, the treatments induced the following major effects: (1) a reduction of more than 99% of the number of *Fusarium oxysporum* colonies in soil; (2) an increase of about 200% in *Trichoderma* spp. in soil; (3) a drastic increase in the plant yield, from 6.9–7.5 g for the control to 23.0–25.5 gr for steam treatments; (4) a high reduction of plants per square meter of 94–95% for *Amaranthus retroflexus*, of 96–99% for *Portulaca oleracea*, of 93–94% for *Chenopodium* spp. and of 86–90% for *Hordeum vulgare*, using both steam and exothermic reactions. Similar results were obtained with steam only. Therefore, our results evidence the potential of this approach to control various soil-borne pathogens. Steam and exothermic reaction are thus promising alternatives to chemical soil disinfection for high-value crops.

basil / disinfection / *Fusarium oxysporum* f.sp. *basilici*, *Sclerotinia minor* / *Ocimum basilicum* L.

1. INTRODUCTION

Soil-borne plant pathogens cause extensive damage to many crops by affecting quality and yield. The concept of soil disinfection consists of eradicating soil-borne pests existing in soil before planting, uniformly to the desired depth, with minimal disturbance of the biological equilibrium and without appreciably affecting chemical or physical soil properties (Katan, 2000).

For years, the most common approach to control of soil-borne pests has been soil fumigation before cropping. Methyl bromide is the most effective soil fumigant, but unfortunately it has negative attributes such as health hazards, environmental pollution, and even potential atmospheric ozone depletion. Increased environmental concern has been a major factor in triggering regulatory restrictions of the use of soil fumigants. The search for non-chemical methods to control soil-borne pathogens has recently intensified ahead of the phase-out of methyl bromide in 2005 (UNEP, 1998). The impending phasing out of this fumigant poses new and unprecedented challenges for the agricultural research community since many major crops, especially in intensive agriculture, have become totally dependent on methyl bromide. The situation is now

changing and the potential of non-chemical methods for pest control is being reassessed in an effort to include them in pest-management programs.

Among physical methods, steaming is still the major tool for soil disinfection. The basic principle is to heat the soil to a temperature that will effectively control all existing soil-borne pests, namely pests, weeds and arthropods. The lethal temperature of all kinds of soil-borne pathogens and weed seeds was established in the 1960s. On the basis of these studies, suggestions for soil steaming were drawn up: a temperature of 70 °C should be maintained for at least 30 min to free the soil from the majority of plant diseases and weeds. However, it has been noted that temperatures above 70 °C, and particularly approaching 100 °C, become detrimental to most soil biota (Bollen, 1969, 1985), and undesirable effects include a shift in the soil's microbial equilibrium, resulting in diminished suppressiveness, i.e. a biological vacuum, increased population of certain pathogens, a detrimental effect on beneficial organisms such as rhizobia and mycorrhizae and microbial antagonists (Dawson and Johnson, 1965; Katan, 1984).

The thermal values achieved in soil with steam treatment change if energy-releasing exothermic reaction chemicals are spread onto the soil prior to treatment, thereby generating a

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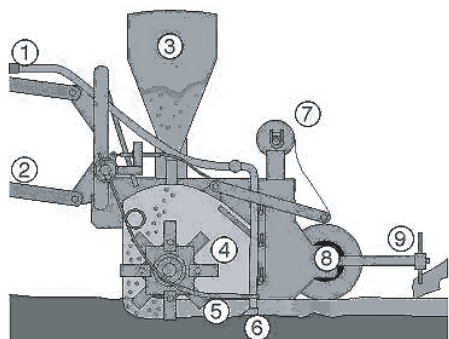


Figure 1. Scheme of the system to perform soil disinfection. (1) Steam feed-pipe; (2) connection arms; (3) hopper of the exothermic reaction product; (4) incorporation into the soil by means of a blade rotor; (5) adjustment of working depth; (6) steam injection; (7) plastic mulch spool; (8) roller; (9) ridging-mulching machine.

temperature peak (“thermal flash”), and the energy released in an exothermic reaction could maintain a useful temperature for controlling pests in the soil during treatment with a mobile steam generator. Moreover, the combination of steam and exothermic reaction could change soil thermal levels through the distribution of temperatures in the soil profile, as well as avoiding a biological vacuum, with minimal concerns with regard to environmental impact or worker safety.

In this article, we describe pre-planting treatments carried out with a self-propelled soil-steaming machine (Fig. 1) designed to release steam after incorporation into the soil of a substance that causes an exothermic reaction (Peruzzi et al., 2003), for the control of two soil-borne pathogens in basil, *Fusarium oxysporum* f.sp. *basilici* and *Sclerotinia minor*, recently described in the United States as the cause of basil stem rot (Koike and O’Brien, 1995). Moreover, the effects of the treatments on microbial population, yield and weed control were evaluated, compared with the use of only steam.

2. MATERIALS AND METHODS

2.1. Experimental design and cultural practices

Experimental plots (120 × 300 cm) were established in open-field conditions at the University of Pisa, Italy in 2003–2005 (thirty-six in 2003–2004 and eighteen in 2005). Split-plot layouts with three replicates were used, preparing different plots in adjacent pieces of land each year. The land was characterized by a sandy soil, with homogeneous properties with no significant difference among the plots before the treatments (62.5% sand; 21.0% silt; 16.5% clay; 1.0% organic matter; pH 7.4; moisture-holding capacity 14.8%). We used uncultivated land from 1999, and soil disinfestation with metham sodium was performed in 2002. The artificial soil inoculations with pathogens were performed in spring each year (April 14, 2003; April 12, 2004; April 11, 2005), and the experimental fields were treated with steam, steam/exothermic reaction chemical and without soil sterilization as control, about a month after inoculation (May 15, 2003; May 14, 2004; May 12, 2005).

Non-inoculated plots were used for yield and weed assessments. A drip irrigation system was established the day after treatments; seedlings of a common Italian variety of basil (*Ocimum basilicum* L. cv. *Genovese gigante*), highly susceptible to *F. oxysporum* f.sp. *basilici* and *S. minor*, were transplanted with 20-cm spacing between the plants two days after soil treatments, 30 plants m⁻². No chemical fertilizers, herbicides or insecticides were applied during the growing season.

2.2. Inoculation with the pathogens

Monosporic isolate of *F. oxysporum* f.sp. *basilici* was obtained from wilted basil plants; each year, infected wheat kernels were produced by adding 100 mL of tap water to 100 g of wheat kernels in a 500-mL flask and autoclaved on two successive days for 20 min at 120 °C and 1.06 kg cm⁻² steam pressure, according to Imolehin et al. (1980) with minor modifications. The autoclaved kernels were inoculated with mycelial disks from 3-day-old cultures of monosporic isolate of *F. oxysporum* f.sp. *basilici*: cultures were aged in the flasks at room temperature (about 20 °C) for 6 weeks before use.

An isolate of *S. minor* was obtained from diseased lettuce plants and sclerotia were produced each year on autoclaved oat seeds using the previous method. The autoclaved oat seeds were inoculated with mycelial disks from 3-day-old cultures of the isolate of *S. minor*: cultures were aged in the flasks at room temperature (about 20 °C) for 12 weeks before use. The sclerotia were recovered by using 1.98 mm (to retain coarse oat debris) and 0.25 mm (to retain sclerotia) sieve sizes. Finally, the sclerotia were dried at room temperature for 24 h and stored at 4 °C until needed.

Artificial soil infestation was carried out about a month prior to application of the soil treatment, to simulate realistic field conditions when basil sowing was performed. Infected wheat kernels were applied to the soil at 20 g m⁻² in three rows of 10 × 300 cm at 8–10 cm depth in 2003–2005, and sclerotia, 15 g m⁻², were applied to the soil in three rows of 10 × 300 cm at 4–6 cm depth in 2004–2005. Propagule counts were performed each year before inoculating the soil to produce similar disease incidences in the control.

2.3. Soil treatments

The equipment used was a self-propelling machine able to inject steam deep down through special feeding devices as it moves (Celli S.p.A., Forlì, Italy).

The machine is able to spread on the soil the exothermic reaction chemical from a hopper, and then it is incorporated into the soil by means of a blade rotor. Behind the rotor, a steam injection system is collocated and connected to the steam generator via a feed – pipe: this steam dispenser can be filled at various depths (in our trials it was set at 15 cm depth) by a system for the adjustment of the rotor working depth. After the release of the steam or steam/exothermic reaction chemical, a plastic film covers the soil by the action of a roller and a ridging – mulching machine. The steam discharge is 600 kg h⁻¹, with a pressure of 1.18 MPa. The injection system is 160 cm wide with 70 holes for steam-dispensing: from 2004 an improved steam dispenser, characterized by different hole profiles, was

designed by Settore Meccanica Agraria of the Dipartimento di Agronomia e Gestione dell'Agroecosistema, University of Pisa (Pisa, Italy) and used.

The treatments performed by the self-propelling machine were steam and steam/potassium hydroxide (KOH) treatments, using 1.000 kg ha⁻¹ of KOH and with a feeding speed of the machine of 60 m h⁻¹. After each treatment the soil was covered with a 40-m-thick black polyethylene film for 24 h, then it was removed. The control plots were uncovered.

2.4. Temperature, disease, microbial and growth assessment, and weed control

Temperatures in the soil profile were monitored in each trial using PT100 sensors that send a voltage signal to data loggers from which data are acquired and recorded on a personal computer using specially designed software. Temperatures were recorded at 0–10 and 11–20 cm depths for 3 h after treatments, reporting the duration of 4 thermal periods (<40 °C, 40–60 °C, 60–80 °C, >80 °C) according to the depth and treatments (data from Settore Meccanica Agraria of the Dipartimento di Agronomia e Gestione dell'Agroecosistema, University of Pisa, Pisa, Italy).

The plant monitoring was performed daily, and disease incidence was evaluated at the end of the experiments by counting the number of healthy and diseased plants (without considering the stage of disease) in the plots, considering all the plants in each plot, for one month after transplanting. Disease evaluation was performed considering all symptomatic plants and re-isolating pathogens from diseased plants, using Komada medium for *F. oxysporum* f.sp. *basilici* and potato dextrose agar (PDA) for *S. minor*. A microscopical identification of pathogens was performed, and 30% of non-symptomatic plants per plot were processed for pathogen isolation. The effectiveness of the treatments on basil was expressed as disease reduction (%) compared with untreated inoculated plots.

The effects of treatment on fungal and actinomycete communities, using steam/KOH or only steam, compared with untreated plots, were evaluated. Soil samples were collected from a plow layer at a depth of 0–10 cm, immediately before planting, from three points in each plot and mixed well to make one composite sample. The composite samples were eased through a 4-mm mesh sieve and kept at 4 °C until microbial analysis. Sub-samples of soil (1 g) suspended in sterile water (10⁻³ w/v) were used to evaluate total fungi, *F. oxysporum*, *Trichoderma* spp. and actinomycetes, sprinkling 1 mL of suspension in petri dishes prepared with 20 mL of selective medium (VDYA for fungi, Komada for *F. oxysporum*, P190 for *Trichoderma* and water-agar for actinomycetes) according to standard methods (Papavizas and Davey, 1959; Komada, 1975; Ho and Ko, 1979). Incubation was conducted at 25 °C and the colonies were counted after seven days. For each soil sample/microbial group, three replicates were used. The results were expressed as colony-forming units (cfu) per g of soil.

In 2004–2006 we assessed the yield of basil in non-inoculated plots, expressed as an increase in fresh weight and dry weight in plots treated with steam/KOH or only steam, compared with untreated plots. The measurement of fresh weight was evaluated by removing all the plants in each plot from the

soil one month after treatment, washing off any loose soil, blotting the plants gently with a soft paper towel to remove any excess surface moisture, cutting off the roots and weighing immediately. The dry weight was performed by drying the plants previously used for determining fresh weight in a oven set at 100 °C overnight, and cooling the plants in a closed plastic bag. The effect of field steaming on weed development was investigated each year in non-inoculated plots, evaluating species and number of weeds m⁻², in order to evaluate the herbicidal effect of the treatments.

2.5. Data analysis

The effects of soil treatments were compared by analysis of variance in a split-plot design. The least significant difference (LSD) at the 5% level was calculated for the comparison of treatments for disease incidence, microbial and growth assessment, and weed control. The differences in temperature among the treatments were analyzed by Duncan's Multiple Range test at the 5% level.

3. RESULTS AND DISCUSSION

3.1. Temperatures

We report the duration of the thermal periods according to depth and treatment, to evaluate the time of permanence of useful temperatures for pest control. Considering the soil at 0–10 cm depth, temperatures were maintained at 40–60 °C for at least 155 min after treatment, in three hours of recording (Tab. I), with no significant differences between the duration of thermal periods according to treatment, although combining steam with KOH determined a longer time of permanence at higher temperatures (40–60 °C) compared with steam only in each year of the trial.

At 11–20 cm depth (Tab. II) we report the duration of thermal periods at a higher range of temperatures compared with 0–10 cm depth, and these results could be due to the position of the steam dispenser in the soil profile (at 15 cm depth). At higher thermal periods (above 60 °C), significant differences

Table I. Duration of the thermal periods at 0–10 cm depth according to treatment (mean data from three replications per treatment).

| Thermal periods | Duration of the thermal periods (min) at 0–10 cm depth | | | | | |
|-----------------|--|-----------|-------|-----------|-------|-----------|
| | 2003 | | 2004 | | 2005 | |
| | Steam | Steam/KOH | Steam | Steam/KOH | Steam | Steam/KOH |
| <40 | 25 a ¹ | 10 a | 16 a | 6 a | 12 a | 7 a |
| 40–60 | 155 a | 170 a | 164 a | 174 a | 168 a | 173 a |
| 60–80 | – | – | – | – | – | – |
| >80 | – | – | – | – | – | – |

¹ Values in the same row (in the same year) followed by the same letter do not differ significantly according to Duncan's Multiple Range test ($P = 0.05$).

Table II. Duration of the thermal periods at 11–20 cm depth according to treatment (mean data from three replications per treatment).

| Thermal periods | Duration of the thermal periods (min) at 11–20 cm depth | | | | | |
|-----------------|---|-----------|-------|-----------|-------|-----------|
| | 2003 | | 2004 | | 2005 | |
| | Steam | Steam/KOH | Steam | Steam/KOH | Steam | Steam/KOH |
| <40 | – | – | – | – | – | – |
| 40–60 | 156 a ¹ | 146 a | 157 a | 146 a | 155 b | 133 a |
| 60–80 | 22 a | 28 b | 20 a | 24 a | 21 a | 32 b |
| >80 | 2 a | 6 b | 3 a | 10 b | 4 a | 15 b |

¹ Values in the same row (in the same year) followed by the same letter do not differ significantly according to Duncan's Multiple Range test ($P = 0.05$).

Table III. Effectiveness of treatments (steam or steam/KOH) on incidence of *F. oxysporum* f.sp. *basilici*, expressed as disease reduction (%) compared with the untreated control (mean data from three replications per treatment).

| Year | <i>F. oxysporum</i> f.sp. <i>basilici</i> / basil | | |
|------|---|-------------------------|-----------|
| | Control | Steam | Steam/KOH |
| 2003 | Diseased plants | 23.0±3.0 a ¹ | 7.3±1.2 b |
| | Disease reduction (%) | – | 69.6 a |
| 2004 | Diseased plants | 25.3±0.6 a | 3.0±0.0 b |
| | Disease reduction (%) | – | 88.1 a |
| 2005 | Diseased plants | 24.3±0.6 a | 2.7±0.6 b |
| | Disease reduction (%) | – | 88.9 a |

¹ Values in the same row followed by the same letter do not differ significantly according to the LSD test ($P = 0.05$); arc sin square root was used prior to % data analysis, non-transformed means are presented.

between treatments are shown during the three years of experimental trials, with a longer duration determined by the use of KOH combined with steam in comparison with only steam.

In every case, the trend of soil temperature registered in all the trials at 11–20 cm depth is characterized by a few minutes at 80 °C or above (a “thermal flash”), then a decrease and staying above 60 °C for at least 20 min, and in the following 133 min or more, the soil temperature is above 40 °C: the use of KOH caused a longer duration of the higher thermal periods, even if in 2004 significant differences by the use of KOH are registered only above 80 °C. Otherwise, at 0–10 cm depth, we report a more homogeneous trend, maintaining temperature in one range (40–60 °C) for almost three hours.

3.2. Management of *F. oxysporum* f. sp. *basilici* and *S. minor*

Symptoms of *Fusarium* wilts became manifest about two weeks after transplanting in each year, appearing as slight vein clearing on the outer portion of the younger basil leaves, followed by epinasty of the older leaves. One month after transplanting, symptomatic plants were stunted, and marginal necrosis of leaves and yellowing of the lower leaves was reported. Observation of vascular tissue was performed at the end of the trial (four weeks after transplanting) and portions of tissue taken from all symptomatic plants and 30% of non-symptomatic plants per plot were used for identification on Komada medium.

The treatments performed reduced the incidence of *Fusarium* wilt (Tab. III), with better results obtained using KOH. In fact, the combination of steam and exothermic reaction chemicals reduced the incidence of disease by 76.9–95.9%, better than using only steam, with a reduction of 69.6–88.9%. The results confirm that a combination of steam and exothermic reaction is effective at reducing disease incidence, even if the improvement of the combination is limited: the major effect is due to the use of steam only, able to reduce the disease by 69.6% or more, while the KOH seems to increase the efficacy by 6.8–7.3%. A different steam dispenser was used from the 2004 trials, optimizing the treatments and obtaining better results: an

Table IV. Effectiveness of treatments (steam or steam/KOH) on incidence of *Sclerotinia minor*, expressed as disease reduction (%) compared with the untreated control (mean data from three replications per treatment).

| Year | <i>S. minor</i> /basil | | |
|------|------------------------|-------------------------|-----------|
| | Control | Steam | Steam/KOH |
| 2003 | Diseased plants | 16.0±1.0 a ¹ | 2.7±0.6 b |
| | Disease reduction (%) | – | 83.1 a |
| 2004 | Diseased plants | 17.7±1.2 a | 2.3±0.6 b |
| | Disease reduction (%) | – | 87.0 a |

¹ Values in the same row followed by the same letter do not differ significantly according to the LSD test ($P = 0.05$); arc sin square root was used prior to % data analysis, non-transformed means are presented.

Table V. Effects of treatments (steam or steam/KOH) on fungal and actinomycete populations, expressed as colony-forming units (cfu) per g of soil (mean data from three replications per treatment).

| Treatment | Total Fungi | Actinomycetes | <i>Trichoderma</i> spp. | <i>F. oxysporum</i> in inoculated plots |
|-----------|-------------------------|---------------|-------------------------|---|
| Control | 28.3±2.1 a ¹ | 55.0±5.0 a | 11.0±1.0 b | 317.7±5.7 a |
| Steam | 31.3±3.1 a | 63.7±5.7 a | 34.3±1.2 a | 1.0±0.0 b |
| Steam/KOH | 31.7±1.5 a | 56.3±4.6 a | 32.7±1.5 a | 1.0±0.0 b |

¹ Values in the same column followed by the same letter do not differ significantly according to the LSD test ($P = 0.05$).

increase of 18.5–19.3% considering steam treatments and of 18.0–19.0% in steam/KOH treatments. No infection by *F. oxysporum* f.sp. *basilici* was registered in non-symptomatic plants.

Symptoms of stem rot caused by *Sclerotinia minor* became manifest about three weeks after transplanting in each year, and the stems developed water-soaked spots which later were covered with a cottony white growth. As the disease progresses, affected portions of the stem develop a bleached appearance, appearing as a browning of the basil stem and epinasty of the leaves with marginal necrosis. One month after transplanting, symptomatic plants were stunted with visible mycelial on the basal part of the stem and on the surface of the soil near the plant. Observation of vascular tissue was performed at the end of the trial (four weeks after transplanting) and portions of tissue taken from symptomatic plants were used for identification.

Steam treatments and the combination of steam/KOH reduced disease incidence caused by *Sclerotinia minor* (Tab. IV). Control of *S. minor* was better with KOH than with steam only, but as shown in the *F. oxysporum* f.sp. *basilici* / basil trials, the beneficial improvement of the combination is limited. In fact, using steam/KOH, we obtained a disease reduction of 93.8–96.1%, compared with 83.1–87.0% obtained with only steam, with an increase in *S. minor* control of 9.1–10.7% caused by the use of the exothermic reaction chemical. No infection by *S. minor* was registered in non-symptomatic plants.

3.3. Effects of treatments on fungal and actinomycete populations

Additional experiments in 2004 revealed no reductions after treatments in the number of total fungi and actinomycetes developed on specific selective medium, without significant differences between treatments. Otherwise, the treatments caused significant reductions in the number of *Fusarium oxysporum* colonies (samples taken from inoculated plots) compared with the untreated control (cfu per g of soil reduction of more than 99%). Moreover, steam/KOH or only steam treatments caused an increase in the number of cfu per g of soil of *Trichoderma* spp. of 197.3% and 211.8%, respectively (Tab. V).

These results show that a mobile steam generator, combined or not with exothermic reaction chemicals, does not cause a biological vacuum, but it is able to increase beneficial biota such as *Trichoderma* spp. and drastically reduce a potential pathogen such as *F. oxysporum*.

3.4. Effects on yield

Steam treatments and the combination of steam and exothermic reaction produced positive effects on yield of basil in non-inoculated plots (Tab. VI). Basil fresh weight increased from 6.9–7.5 g to 23.0–25.3 g in steam treatments and to 23.7–25.5 g in steam/KOH treatments, while basil dry weight increased from 1.5–1.7 g to 4.8–5.1 g in steam treatments and to 5.0–5.1 g in steam/KOH treatments.

Therefore, the treatments drastically increased plant fresh and dry weights in all the trials, but the exothermic reaction chemical did not cause significant differences compared with the use of steam only during all three years of trials.

3.5. Weed control

The effects of treatments on main weed development are shown in Table VII. The field used in the reported trials was naturally infested by various weeds, such as *Amaranthus retroflexus*, *Portulaca oleracea*, *Hordeum vulgare*, *Chenopodium* spp. and other species. The results achieved were very interesting, with a satisfactory weed control: in fact, using steam/exothermic reaction caused a significant reduction in plants m⁻² in *Amaranthus retroflexus* (93.9–94.7%), *Portulaca oleracea* (96.1–98.9%), *Chenopodium* spp. (92.7–93.8%) and *Hordeum vulgare* (85.6–90.0%), even if no statistical differences between steam treatment and steam/KOH treatment are reported.

4. CONCLUSION

To improve the efficacy of a soil-steaming treatment characterized by a short period of exposure to the required

Table VI. Effects of treatments (steam or steam/KOH) on yield of basil as fresh weight or dry weight (g) compared with untreated controls (mean data from three replications per treatment).

| | | Basil productivity | | |
|------|--------------|------------------------|------------|------------|
| Year | | Control | Steam | Steam/KOH |
| 2003 | Fresh weight | 6.9±0.4 a ¹ | 23.0±4.5 b | 23.7±5.5 b |
| | Dry weight | 1.5±0.3 a | 5.0±1.0 b | 5.1±0.9 b |
| 2004 | Fresh weight | 7.0±0.6 a | 23.2±5.4 b | 24.9±6.6 b |
| | Dry weight | 1.6±0.2 a | 4.8±1.3 b | 5.1±1.5 b |
| 2005 | Fresh weight | 7.5±0.9 a | 25.3±4.4 b | 25.5±3.6 b |
| | Dry weight | 1.7±0.3 a | 5.1±1.5 b | 5.0±1.6 b |

¹ Values in the same row followed by the same letter do not differ significantly according to the LSD test ($P = 0.05$).

Table VII. Effects of treatments (steam or steam/KOH) on weed control expressed as plants m⁻², compared with untreated controls (data from three replications per treatment).

| Year | Weed (plants m ⁻²) | Weed control | | |
|------|--------------------------------|---------------------------|-------------|-------------|
| | | Control | Steam | Steam/KOH |
| 2003 | <i>Amaranthus retroflexus</i> | 583.3±29.0 a ¹ | 96.0±11.5 b | 35.3±10.0 b |
| | <i>Portulaca oleracea</i> | 97.7±10.8 a | 0.0±0.0 b | 2.0±1.0 b |
| | <i>Chenopodium</i> spp. | 582.3±16.6 a | 25.0±1.0 b | 39.7±2.1 b |
| | <i>Hordeum vulgare</i> | 67.0±3.5 a | 5.0±3.0 b | 9.0±1.0 b |
| | Other | 3.1±1.3 a | 1.0±0.0 a | 2.1±1.1 a |
| 2004 | <i>Amaranthus retroflexus</i> | 560.5±25.1 a | 80.1±10.0 b | 30.2±6.0 b |
| | <i>Portulaca oleracea</i> | 90.1±10.8 a | 2.1±1.0 b | 1.0±1.5 b |
| | <i>Chenopodium</i> spp. | 452.5±10.3 a | 20.2±1.5 b | 31.3±1.1 b |
| | <i>Hordeum vulgare</i> | 60.1±3.0 a | 6.0±2.0 b | 6.0±4.2 b |
| | Other | 4.2±1.1 a | 2.0±1.0 a | 2.2±1.0 a |
| 2005 | <i>Amaranthus retroflexus</i> | 550.5±20.0 a | 75.0±8.4 b | 28.7±4.3 b |
| | <i>Portulaca oleracea</i> | 80.2±9.8 a | 1.0±0.0 b | 3.1±1.5 b |
| | <i>Chenopodium</i> spp. | 500.5±15.3 a | 21.5±1.3 b | 36.4±2.2 b |
| | <i>Hordeum vulgare</i> | 62.2±4.4 a | 4.5±2.1 b | 7.1±2.1 b |
| | Other | 2.5±1.5 a | 1.5±1.5 a | 2.0±1.0 a |

¹ Values in the same row followed by the same letter do not differ significantly according to the LSD test ($P = 0.05$).

temperature, we combined exothermic reaction chemicals with steam to achieve a slightly higher treatment temperature, and to change soil thermal levels through the distribution of temperatures in the soil profile, as well as to avoid dramatic shifts in the soil's microbial equilibrium. The experimental trials showed that the combination of steam and exothermic reaction chemicals reduced the incidence of *F. oxysporum* f.sp. *basilici* and *S. minor* in basil, with better results compared with the use of only steam, even if the improvement is limited: in fact, the major effect is due to the use of steam only, able to reduce *Fusarium* disease by 69.6% or more and *S. minor* disease by 83.1%, while the KOH seems to increase the efficacy by 6.8–7.3% for *Fusarium* and by 9.1–10.7% for *S. minor*. Therefore, the use of exothermic reaction chemicals, even if they have positive effects on disease control, need to be evaluated considering the cost of application in relation to the level of inoculum in the soil, and considering the incremental control of soil inoculum that could happen in repeated treatments on the same land. The 2002 results on microbial assessment (Triolo et al., 2004) were confirmed in the 2004 trials, with a dramatic reduction in the number of *Fusarium oxysporum* colonies (cfu per g of soil reduction of more than 99%) and with minimal effects on total fungi and actinomycetes, while the number of *Trichoderma* cfu per g of soil was increased by about 200%. Thus, one of the main concerns about steam treatments, biological vacuum, seems to be limited, preserving or increasing the population of various microorganisms and decreasing potential pathogens. The effects of treatment on weeds are also encouraging, with a reduction in plants m⁻² of above 85.6% for the four weeds monitored during the trials, according to the results obtained on seed germination (Peruzzi et al., 2004). Moreover, dramatic increases in yield are reported: in a basil plant treated with steam/KOH or steam only there was an increase in fresh weight of above 300%, and this positive effect

is confirmed by the dry weight. The use of a combination of KOH with steam is safe and does not cause hygiene-toxicological or environmental issues, but generally has undesirable agronomic effects, such as an increase in soil pH, exchangeable K (KOH) and conductivity. If a low amount of exothermic reaction chemicals is used, the variations are mild and disappear in 15 to 30 days after treatment (Mazzoncini et al., 2002). To conclude, the use of exothermic reaction chemicals seems to be useful for controlling pathogens, with a constant increase in disease control in all the experiments, even if the major effects were caused by steam treatments, in particular considering microbial assessment, yield and weed control. Thus, based on our research with soil steaming, the exothermic reaction can improve the effectiveness of treatment carried out by mobile steam generators, adopting a short treatment exposure time, improving the steam-based method. Our conclusion is, therefore, that a combination of steam and exothermic reaction can be included in integrated pest-management programs.

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