

# Evaluation of Broadcast Steam Application with Mustard Seed Meal in Fruiting Strawberry

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**Abstract.** Soil disinfestation with steam has potential to partially replace fumigants such as methyl bromide, chloropicrin, and 1,3-dichloropropene because it is effective, safer to apply, and has less negative impact on the environment. Here, we compared the efficacy of steam and steam + mustard seed meal (MSM) to chloropicrin on soil disinfestation, plant growth, and fruit yield in a strawberry (*Fragaria × ananassa*) fruiting field. The MSM was applied at 3368 kg·ha<sup>-1</sup> before the steam application. Steam was injected into a 3-m-wide reverse tiller that was set to till 30 to 40 cm deep. Soil temperatures at depths of 10, 20, 25, and 35 cm were monitored. Steam and steam + MSM treatments reduced the viability of purslane seeds and nutsedge tubers, microsclerotia density of *Verticillium dahliae*, propagule density of *Pythium ultimum*, cumulative weed densities, and biomass compared with the nontreated control. Moreover, the steam application was as efficacious as chloropicrin on these pests. The growth and fruit yield of strawberries grown on soils previously treated with the steam and steam + MSM treatments were similar to those in the chloropicrin treatment and were higher than those in the nontreated control. Our study indicated that steam, steam + MSM, and chloropicrin are equally effective at suppressing weeds and soilborne pathogens. These results suggest that the steam and steam + MSM treatment can be a practical alternative for soil disinfestation in conventional and organic strawberry fields.

Methyl bromide (MB) has been widely used for decades as a soil fumigant in conventional strawberry fields (Fennimore and Goodhue, 2016). But MB has been linked to depletion of the atmospheric ozone layer and its use has largely been phased out (Sen et al., 2010). Chloropicrin alone and in combination with 1,3-dichloropropene (1,3-D) serves as the primary replacement for MB (Moldenke and Thies, 1996; South et al., 1997). Fumigant use regulations in California, however, require buffer zones near sen-

sitive sites like hospitals and schools, which complicates the fumigation of agricultural fields (Goodhue et al., 2016). Beauvais (2012) and Lewis (2012) reported that chloropicrin causes transient mucous membrane and eye irritation at relatively low concentrations, and every effort is made to reduce bystander exposure to fumigants. Therefore, there is much interest in fumigant alternatives for use in areas that cannot be fumigated, such as sensitive sites or organic fields.

Among the nonfumigant alternatives, anaerobic soil disinfestation (ASD) is the most promising so far (Shennan et al., 2018), but ASD requires use of plastic film, which creates solid waste and high nitrogen carbon sources like rice bran, which can pollute groundwater (Messiha et al., 2007). In contrast, soils disinfestation with steam has less impact on the environment and is safer to applicators and bystanders than chemical fumigants, which pose risk of exposure (Fennimore and Goodhue, 2016; Kim et al., 2020). Moreover, Fennimore et al. (2014) reported steam to be a dependable nonfumigant method of soil disinfestation for control of pathogens and weeds with similar efficacy as MB in strawberry fruit production fields.

Steam coapplication with MSM may be a means to enhance performance so that the application can be made faster and more efficiently than steam alone. MSM may reduce infection by plant pathogens such as *Rhizoctonia solani* (Mazzola et al., 2007), *Pythium abipressorium* (Weerakoon et al., 2012), and *Cylindrocarpum* and *Rhizoctonia* spp. (Mazzola and Brown, 2010), and may control weeds like *Lolium multiflorum* and *Stellaria media* (Boydston and Anderson, 2008; Handiseni et al., 2011). Borek and Morra (2005) and Brown et al. (1991) found that MSM contains glucosinolates that undergo enzymatic hydrolysis to thiocyanates, isothiocyanates, nitriles, and other compounds in moist soil. Among the hydrolyzed compounds, radial growth suppression of *Helminthosporium solani* and *Verticillium dahliae* were correlated to allyl isothiocyanate (AITC) concentration, which is a form of volatile isothiocyanate (Olivier et al., 1999; Petersen et al., 2001). According to Dai and Lim (2014), AITC was released from MSM up to 13 mg·g<sup>-1</sup>. AITC is available as a natural product in MSM or in synthetic form as Dominus (99.8% AITC; Isagro USA Inc., Morrisville, NC) (Kim et al., 2019, 2020). Previous works have shown that Brassicas incorporated into soil can enhance efficacy of solarization (Stapleton et al., 2000). Our hypothesis is that steam plus MSM would improve pest control efficacy compared with either treatment alone.

Steam kills weed seeds and pathogens with high temperature. Previous studies indicated that steam application must raise soil temperatures to 60 °C in the 0 to 15-cm layer for most vegetables to control weeds and 60 °C to a depth of 35 cm for strawberry to control weeds and pathogens (Baker, 1962; Fennimore et al., 2014). Steam efficacy on soil pests has been typically evaluated by weed densities and biomass, the pathogen propagule viability, diseased plant counts, plant size, and fruit yield (Fennimore et al., 2014; Kim et al., 2020; Samtani et al., 2012).

The objectives of this project were to evaluate efficacy of steam and steam + MSM applications for soil disinfestation, plant growth enhancement, and fruit production.

## Materials and Methods

**Steam application.** Field trials were established at the Spence U.S. Department of Agriculture Agricultural Research Service research farm near Salinas, CA, on a sandy loam soil. Treatments included 1) nontreated control, 2) MSM (Farm Fuel, Watsonville, CA) alone 3368 kg·ha<sup>-1</sup>, 3) steam alone, 4) steam + MSM 3368 kg·ha<sup>-1</sup>, and 5) chloropicrin (Trical, Hollister, CA). The MSM was applied before the steam application with a fertilizer spreader. HOB0 data loggers (U12 Outdoor; Onset Computer Corp., Pocasset, MA) were used to monitor soil temperatures at depths of 10, 20, 25, and 35 cm in the nontreated control, steam, and steam + MSM treatments. Treatments were replicated four

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times and arranged in a randomized complete block design. Steam was applied on 14 Sept. 2018 and 6 Sept. 2019. The field-scale steam applicator (Southern Turf Nurseries, STN, Elberta, AL) was equipped with a 300-hp diesel-fueled Cleaver Brooks steam generator mounted on a trailer towed by a tractor (Fig. 1). Steam was injected into a 3-m-wide reverse tiller (Northwest Tillers, Yakima, WA) that was set to till 30 to 40 cm deep immediately blending steam into the soil. Following steam application, beds were shaped and mulched with plastic film, and chloropicrin was applied through the drip irrigation system on 18 Oct. 2018 at a rate of 28.2 g·m<sup>-2</sup> and on 17 Oct. 2019 at a rate of 24.2 g·m<sup>-2</sup>. Strawberry plants (*Fragaria × ananassa* cv. Cabrillo) were transplanted 7 Nov. 2018 and 15 Nov. 2019. Fertilizer containing nitrogen, phosphate, potash, and micronutrient was applied via the drip system 11 times from Mar. to Aug. 2019 and 12 times from Feb. to Sept. 2020. The insect and phytopathogen management after transplanting was conducted by suction devices (bug-vacs); dust control and hand weeding; removal of ripe and decayed fruits, infected and dead plants, and plant debris; and proper irrigation to prevent water stress.

**Measurement of plant growth and fruit yield.** The plant vigor was scored from 0 (dead) to 10 (vigorous) per each plant (n = 50). The plant diameters were measured in two dimensions (n = 10). The plant vigor and diameters were measured three times from January to April. Strawberry fruit was harvested 31 times from Apr. to Aug. 2019 and 36 times from Apr. to Sept. 2020 from four



Fig. 1. The mobile applicator (Southern Turf Nurseries, STN, Elberta, AL) used in this test.

replications of 26 to 30 plants in 1.2 m × 6 m (width × length) sample areas. Fruit fresh weight was measured by commercial harvest crews once or twice weekly. Marketable fruit had the following characteristics: more than 10 g in weight; free of mechanical, insect, or disease damage; a minimum of 80% red surface; and attached calyx.

**Bioassay.** *Verticillium dahliae* samples of 25 g of infected soil and 100 common purslane (*Portulaca oleraceae* L.) seeds and 25 yellow nutsedge (*Cyperus esculentus* L.) tubers were contained in nylon mesh bags. The bags were buried 15 mm deep and HOBO data loggers were installed next to the bags immediately after the steam applicator passed. Samples and data loggers were collected after 2 weeks and 3 d, respectively. Following sample retrieval, *V. dahliae* samples were air-dried, hand crushed, and the number of microsclerotia per gram of soil was assessed via the dry plating method on NP-10 semiselective medium according to Kabir et al. (2004). *Pythium ultimum* levels were assessed in pre- and posttreatment soil samples using a wet plating method on semi-selective medium (Klose et al., 2007). The soil samples of pre- and posttreatment were collected 3 d before and after the treatment, respectively. The purslane seed samples were tested for viability using the methods described by Cottrell (1947). A 0.1% (v/v) solution of 2,3,5-triphenyl-tetrazolium chloride (Sigma, St Louis, MO) was used to stain the seeds from the recovered containers and probes. The purslane seed samples were plated on germination paper in petri dishes, cut in half, stained, and kept in the dark at 24 °C for 24 h. The viability of the individual seeds was evaluated under a microscope, according to their staining intensity. Seeds with little or no stain were counted as dead and seeds with obvious stain intensity were counted as live. Weed densities and biomass of resident field weeds in season were assessed at monthly intervals between January and April.

**Statistical analyses.** To compare the treatment effects on viability of purslane seeds,

nutsedge tubers, *V. dahliae*, and *P. ultimum* levels, the nonparametric Kruskal-Wallis tests were used to avoid the assumptions of normality and homogeneity of variance. For pairwise comparisons, the pairwise Wilcoxon rank sum tests were implemented (Hollander and Wolfe, 1973). To respect a fixed significance level in the multiple comparisons ( $\alpha = 0.05$ ), *P* values were adjusted using the correction method suggested by Benjamini and Hochberg (1995). Similar analyses were performed for cumulative weed density and biomass. To compare plant vigor and plant canopy, the mean separation was performed using Duncan's multiple range test, and their estimated mean was graphically presented. These analyses were performed using SPSS 20.0 for Windows (SPSS Inc., Chicago, IL) and R version 4.0.2 (R Core Team, 2019).

The marketable fruit yield was compared among treatments using a mixed-effect model. The mixed-effect model considered both random effects due to observing the same experimental unit over time and fixed effects due to treatment effects. Under the model the rate of expected cumulative yield of each treatment was compared with the nontreated control and among the treatments. For a graphical demonstration, the cumulative total and marketable fruit yield were plotted with respect to the time since the beginning of harvest season (15 Apr. 2019 for the first season and 27 Apr. 2020 for the second season). These analyses were performed by R version 4.0.2 (R Core Team, 2019).

## Results

**Soil temperature.** Time above 60 °C during the first hour in both steam treatments in the 2018–19 trial was 58.5, 58.5, and 59.5 min at the 10, 20, and 25 cm depths, respectively (Table 1). Time above 60 °C during the first hour in the steam-alone treatment in the 2019–20 trial was 59.5, 59.5, and 60.0 min at 10, 20, and 25 cm depths, respectively. Time above 60 °C at 10- and

Table 1. The duration of the soil temperature for the first hour after steam application.

	Treatment	Depth (cm)	60 to 65 °C	65 to 70 °C	>70 °C
			Time (min)		
2018–19	Steam alone	10	9	33.5	16
		20	1.5	57	0
		25	36	23.5	0
		35	0	0	0
	Steam + MSM	10	0.5	1	57.5
		20	0.5	1	58
		25	0	0.5	59
		35	0	0	0
2019–20	Steam alone	10	0	0.5	59
		20	0	0	59.5
		25	0	0.5	59.5
		35	0	0	0
	Steam + MSM	10	0.5	6	53.5
		20	0	34	26
		25	28.5	2	0
		35	0	0	0

The data are means of four replications.  
MSM = mustard seed meal.

20-cm depths in steam + MSM treatment was almost the same as that in the steam-alone treatment except at 25-cm depth where time above 60 °C was 30.5 min (Table 1).

**Pest control.** Steam, steam + MSM, and chloropicrin reduced viability of purslane seed, nutsedge tubers, and *V. dahliae* (Table 2) compared with the nontreated control and MSM alone. Similar patterns were shown for *P. ultimum* levels (Table 3); however, steam, steam + MSM, and chloropicrin were not different because *P. ultimum* levels were low in the nontreated control and MSM alone in the 2019–20 trial. The pest control efficacy of the steam treatment was similar to chloropicrin.

Weeds present were California burclover (*Medicago polymorpha*) and common groundsel (*Senecio vulgaris*) in the 2018–19 trial and burning nettle (*Urtica urens*), chickweed (*Stellaria media*), henbit (*Lamium amplexicaule*), lesser swinecress (*Lepidium didymum*), and perennial ryegrass (*Lolium perenne*) in the 2019–20 trial. The multiple comparisons in the 2018–19 trial showed that there was no significant difference between treatments at  $\alpha = 0.05$ . The multiple comparisons in the 2019–20 trial showed that the effects of the steam alone, steam + MSM, and

chloropicrin had lower weed densities and fresh weights compared with the nontreated control and MSM alone (Table 4).

**Plant growth.** The vigor estimates and plant diameter measurements indicated that strawberry plants in the steam + MSM treatment in the 2018–19 trial were larger and more vigorous than those in the other treatments. Strawberry plants in the steam alone, steam + MSM, and chloropicrin treatments in the 2019–20 trial were greater than those in the nontreated control and MSM alone (Figs. 2 and 3). Strawberry plants in all treatments grew rapidly between March and April of the 2018–19 trial and February and March of the 2019–20 trial (Fig. 3). The plant diameter in the steam and steam + MSM treatments in the 2019–20 trial, especially, grew faster than the nontreated control and MSM treatment. The plants in the steam, steam + MSM, and chloropicrin treatments were larger than the nontreated control and MSM alone treatment during both seasons.

**Fruit yield.** Marketable fruit yield in the steam, steam + MSM, and chloropicrin treatments was higher than that in the nontreated control (Table 5). The table provides the rate of cumulative fruit yield (grams per plant) over time as estimated by the mixed-effect

model for each treatment relative to the nontreated control (i.e., “+” means a faster rate of cumulative yield relative to the nontreated control). Marketable yield in MSM alone was as effective as steam alone during the 2018–19 season, but the MSM alone application was not as effective as steam alone in the 2019–20 season. Moreover, the yield in the steam + MSM was similar to that in the chloropicrin treatment during both seasons (Table 5, Fig. 4). The estimated rate of cumulative fruit yield was similar among the steam alone, steam + MSM, and chloropicrin in the 2019–20 season.

## Discussion

Soil disinfestation with steam raises fewer concerns about exposure to fumigants (Fennimore et al., 2014; Kim et al., 2019, 2020; Samtani et al., 2012). van Loenen et al. (2003) and Melander and Jørgensen (2005) demonstrated that steam applied to the soil raising temperatures to 60 °C for 3 min, reduced *V. dahliae* and *P. ultimum* and weed emergence in the laboratory. Steam injected and mixed with soil reaching 70 °C for 20 min controlled soilborne pathogens and weeds in California strawberry fields

Table 2. Viability of purslane seeds and nutsedge tubers and microsclerotia density of *Verticillium dahliae*.

Treatment	Purslane		Nutsedge	<i>V. dahliae</i>	
	2018–19	2019–20	2019–20	2018–19	2019–20
	% Viable			Microsclerotia/g	
Nontreated control	77.5 ± 9.9 b	47.3 ± 5.2 b	83.5 ± 6.7 b	687.7 ± 114.0 b	220.0 ± 69.4 b
MSM alone	85.5 ± 3.9 b	59.3 ± 7.0 b	77.0 ± 10.4 b	552.2 ± 80.2 b	177.8 ± 71.7 b
Steam alone	3.0 ± 3.0 a	0.0 ± 0.0 a	7.0 ± 6.4 a	1.2 ± 1.2 a	0.3 ± 0.2 a
Steam + MSM	0.5 ± 0.5 a	0.0 ± 0.0 a	0.0 ± 0.0 a	0.2 ± 0.2 a	0.0 ± 0.0 a
Chloropicrin	2.1 ± 2.1 a	0.0 ± 0.0 a	1.5 ± 1.5 a	0.2 ± 0.2 a	2.5 ± 0.6 a

The data are means of four replications.

The lowercase letters indicate significantly different treatment effects at the significance level of  $\alpha = 0.05$ .

MSM = mustard seed meal.

Table 3. Propagule density of *Pythium ultimum* before and a week after treatment.

Treatment	2018–19			2019–20		
	Pretreatment	Posttreatment	% Reduction	Pretreatment	Posttreatment	% Reduction
	Propagule (n.g <sup>-1</sup> )			Propagule (n.g <sup>-1</sup> )		
Nontreated control	26.0 ± 3.0	27.7 ± 6.6 b	–7	3.7 ± 2.1	3.0 ± 2.6	19
MSM alone	26.7 ± 4.7	30.0 ± 14.3 b	–12	1.7 ± 0.6	1.7 ± 0.8	0
Steam alone	32.0 ± 8.0	0.0 ± 0.0 a	100	2.7 ± 0.0	0.0 ± 0.0	100
Steam + MSM	47.7 ± 17.7	0.0 ± 0.0 a	100	2.3 ± 1.0	0.0 ± 0.0	100
Chloropicrin	29.3 ± 4.3	0.7 ± 0.0 a	98	2.0 ± 0.9	0.0 ± 0.0	100

The data are means of four replications.

The lowercase letters indicate significantly different treatment effects at the significance level of  $\alpha = 0.05$ .

MSM = mustard seed meal.

Table 4. The cumulative season-long weed densities and biomass.

Treatment	Weed density		Fresh wt	
	2018–19	2019–20	2018–19	2019–20
	No./m <sup>2</sup>		g·m <sup>-2</sup>	
Nontreated control	1.38 ± 0.09	2.42 ± 0.66 b	4.44 ± 0.87	5.60 ± 1.72 b
MSM alone	1.53 ± 0.14	0.85 ± 0.11 b	8.55 ± 1.89	2.49 ± 0.43 b
Steam alone	0.75 ± 0.07	0.08 ± 0.03 a	1.95 ± 0.26	0.28 ± 0.13 a
Steam + MSM	1.16 ± 0.17	0.11 ± 0.03 a	3.49 ± 0.64	1.26 ± 0.58 a
Chloropicrin	0.99 ± 0.18	0.06 ± 0.02 a	4.19 ± 1.55	0.10 ± 0.09 a

The data are means of four replications.

The lowercase letters indicate significantly different treatment effects at the significance level of  $\alpha = 0.05$ .

MSM = mustard seed meal.

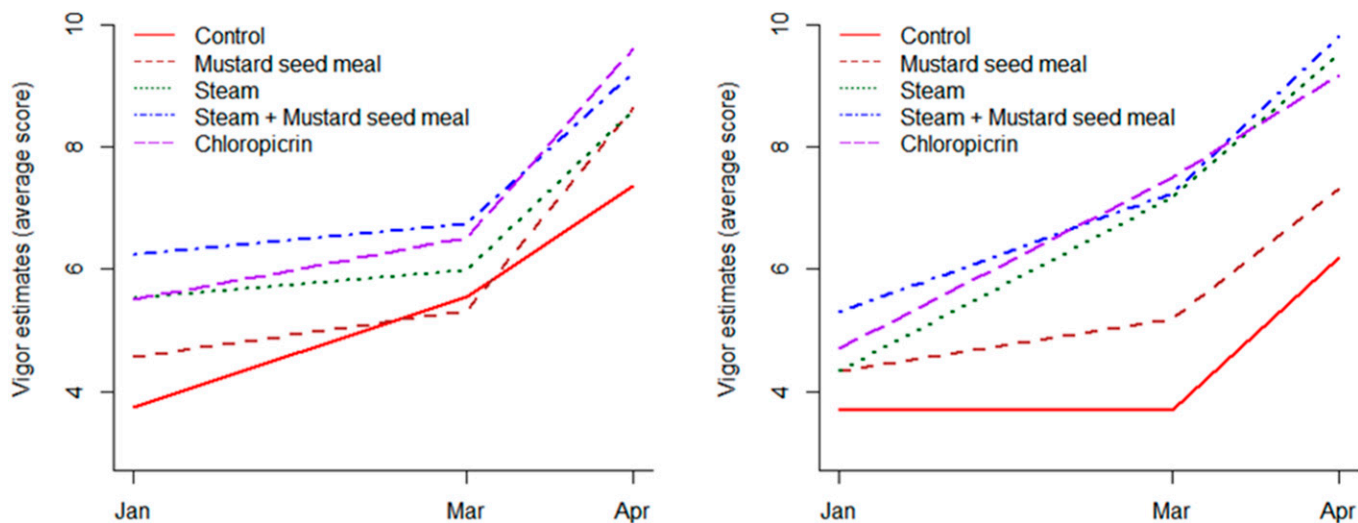


Fig. 2. Vigor estimates of strawberry plants in 2019 (left) and 2020 (right). The data are means of four replications.

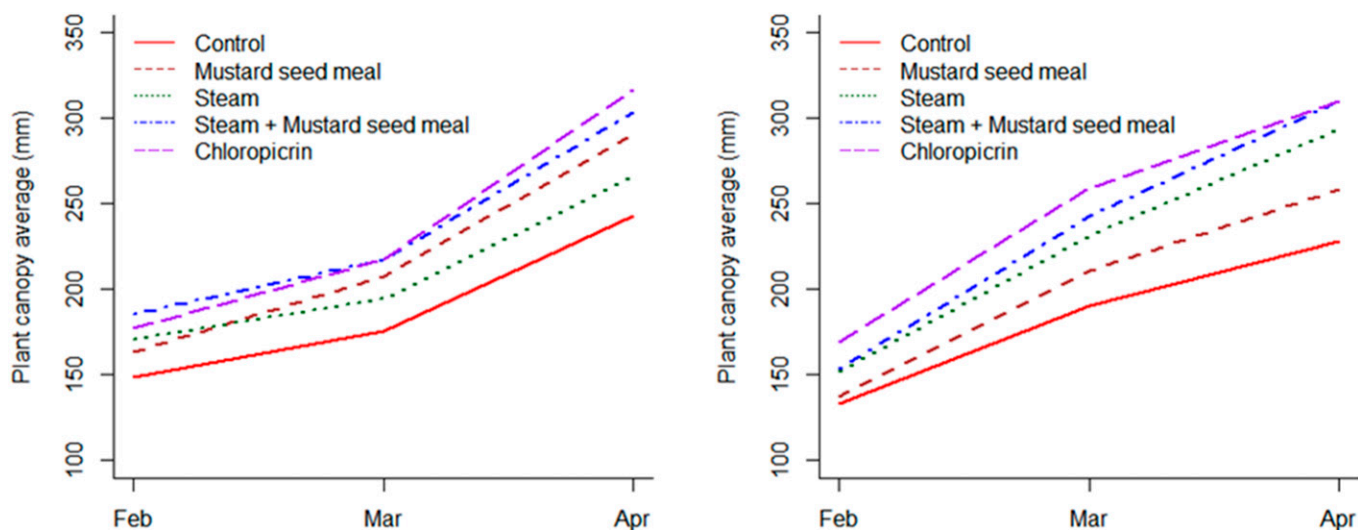


Fig. 3. Strawberry plant diameter during periods of rapid growth in 2019 (left) and 2020 (right). The data are means of four replications.

Table 5. The estimated slopes in Fig. 3 and *P* values calculated under the mixed effects models (“+” means relative to the nontreated control).

Yr	Treatment	Marketable fruit yield	
		Estimate	
		Avg daily cumulative grams per plant	<i>P</i> value
2019	Nontreated control	27.5	
	MSM alone	+6.5	0.028
	Steam alone	+5.5	0.055
	Steam + MSM	+10.7	0.001
	Chloropicrin	+9.1	0.004
2020	Nontreated control	23.5 a	
	MSM alone	+2.3 a	0.357
	Steam alone	+14.2 b	<0.001
	Steam + MSM	+15.8 b	<0.001
	Chloropicrin	+14.8 b	<0.001

The data are means of four replications.

The lowercase letters indicate significantly different treatment effects at the significance level of  $\alpha = 0.05$ .

MSM = mustard seed meal.

(Fennimore et al., 2014; Samtani et al., 2012). Most weeds that emerge are from the shallow layers of the soil. Benvenuti et al. (2001) found that weeds did not emerge from more than 12 cm deep and Pullman et al.

(1981) found that most viable propagules of *V. dahliae* and *Pythium* spp. were in the upper soil layer with fewer propagules at 15 to 30 cm soil depths. Successful field steam application requires an understanding of two im-

portant factors: 1) steam does not disperse far from the injection point in soil, steam only goes where placed, and thus, placement of steam is very important; and 2) soil temperatures following steam application can vary



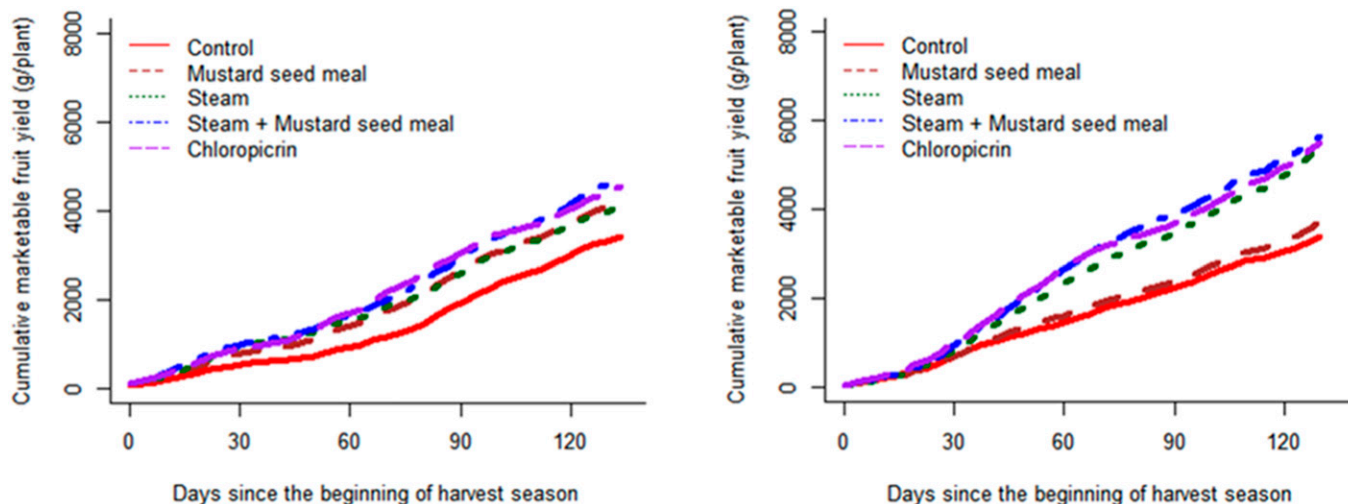


Fig. 4. The cumulative marketable fruit yield in 2019 (left) and 2020 (right). The data are means of four replications.

depending on soil humidity, air temperature, and soil compaction (Kim et al., 2020). Steam was injected into soil at temperatures of 120 °C and depth of tillage was 40 cm. Soil temperatures remained above 60 °C for about 1 h following steam application and penetrated to 25 cm deep (Table 1). The soil temperatures observed in this research, therefore, were adequate to reduce phytopathogens and weed seed viability as well as to protect the crop rooting zone (Fennimore et al., 2014; Samtani et al., 2011).

Applications of steam alone and steam + MSM decreased the viability of purslane seeds, nutsedge tubers, and *V. dahliae*, *P. ultimum* levels, and cumulative weed densities and biomass comparable to chloropicrin (Tables 2–4). However, MSM did not control soil pathogens as well as steam alone and steam + MSM. Angus et al. (1994) reported that pest control by glucosinolate containing shoot and root tissues of *Brassica* species resulted from isothiocyanates (ITCs). Among the ITCs, AITC, biologically active hydrolysis products of glucosinolates, was effective in reducing soil pests (Bangarwa et al., 2011; Brown and Morra, 1997). The volatile nature of AITC aids in its uniform distribution in soil. Moreover, Kim et al. (2020) found that steam + AITC has a complementary effect on pests because steam application increases the mobility of AITC. Therefore, to boost the pesticidal activity of MSM, it is necessary to find methods to increase hydrolysis of glucosinolates (Olivier et al., 1999). On the other hand, the addition of MSM improved growth (Figs. 2 and 3) and fruit yield (Table 5, Fig. 3) of the strawberry plants. This growth may be due to the 4.5% nitrogen contained in MSM (www.farmfuelinc.com/products/). Fennimore et al. (2014) found that ammonium and nitrate level in the soil of steam + MSM treatment was higher than that in nontreated and steam-alone treatments due to the nitrogen in MSM. Dai and Lim (2014) found that MSM contained 33.6% protein, 16.6% lipid, and 5.5% carbohydrate. The nitrogen could be absorbed by plants in the form of

ammonium and nitrate to promote the growth and development. Although MSM increased the plant growth and fruit yield of strawberry due to the nutrients, our data may suggest that MSM alone can be a problem (Tables 2–4).

Fennimore et al. (2014) and Samtani et al. (2011) found that strawberry fruit yields in soils treated with steam were higher than those of nontreated control. Our results also showed that plants grown on soils previously treated with steam produced yields similar to chloropicrin and greater than those of the nontreated controls (Table 5, Fig. 4). The vigor of strawberry plants, moreover, are important because it is linked to fruit yield (Fennimore et al., 2014; Singh et al., 2008). Salamé-Donoso et al. (2010) reported that the diameter of strawberry plants displayed a positive correlation with fruit production regardless of cultivars and production system. The results demonstrated that the strawberry plants in the steam and steam + MSM treatments produced similar quantities of fruit as plants grown in soils previously treated with chloropicrin.

## Conclusion

The finds here suggest the following: 1) pest suppression by steam was similar to that of chloropicrin, 2) MSM did not improve performance of the steam application because steam alone is very effective, 3) strawberry plant vigor and yield responses to the steam application were similar to those to chloropicrin, and 4) MSM might provide nitrogen, although fertilizer benefits from MSM were not directly evaluated in this study. However, despite a decade of favorable data with soil steaming on open field applications, a commercially available technology for 35-cm-deep soil steaming has not been developed yet. There are small or shallow steaming machines on the market. The challenge is to find an economic method of steam application at field scale that can treat large acreages in a timely and cost-effective manner. Cooperative efforts are needed to

bring together interested parties that seek to develop soil steaming technologies for use in conventional and organic fields (e.g., Agricultural Soil Steaming Association, USA, www.soilsteamingassociation.org/).

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