Effects of Short-Term Biosolarization Using Mature Compost and Industrial Tomato Waste Amendments on the Generation and Persistence of Biocidal Soil Conditions and Subsequent Tomato Growth

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ABSTRACT: Conventional solarization and biosolarization with mature compost and tomato processing residue amendments were compared with respect to generation of pesticidal conditions and tomato (*Solanum lycopersicum* L.) plant growth in treated soils. Soil oxygen depletion was examined as a response that has previously not been measured across multiple depths during biosolarization. For biosolarized soil, volatile fatty acids were found to accumulate concurrent with oxygen depletion, and the magnitude of these changes varied by soil depth. Two consecutive years of experimentation showed varying dissipation of volatile fatty acids from biosolarized soils post-treatment. When residual volatile fatty acids were detected in the biosolarized soil, fruit yield did not significantly differ from plants grown in solarized soil. However, when there was no residual volatile fatty acids in the soil at the time of planting, plants grown in biosolarized soil showed a significantly greater vegetation amount, fruit quantity, and fruit ripening than those of plants grown in solarized soil.

KEYWORDS: fumigation alternative, integrated pest management, food waste valorization, tomato waste, soil oxygen content, tomato plant physiology

INTRODUCTION

Alternatives to soil fumigation are gaining interest in order to mitigate the negative environmental and human health effects associated with conventional fumigants.[1] With the phase-out of methyl bromide, the once prominent fumigant heralded for its broad spectrum efficacy but later discovered to contribute to ozone layer depletion,[2] other fumigants such as chloropicrin and 1,3-dichloropropene have gained popularity. However, they present their own toxicity concerns and lack the broad spectrum pesticidal activity of methyl bromide.[3,4] Solarization is an alternative to fumigation and other chemical pesticides that has been adopted in commercial agriculture in various regions worldwide.[1,5] Solarization employs passive solar heating, which is induced by covering moist soil with transparent plastic tarps, to promote thermal inactivation of soil-borne pathogens and remediation of pesticides.[1,6] Studies have shown pest suppression and benefits to tomato crop growth as a result of solarization.[7−10] However, more widespread adoption of solarization faces notable challenges, such as the need for long treatment times, a strong reliance on local climate and weather, and the inability to heat deep levels in soil where certain pests may reside.[1,11]

For instance, solarization research in Southern Italy used extended solarization periods (79, 37, and 34 days) to control weeds and nematodes and to improve crop yields for greenhouse tomatoes and melons. It has been noted that the treatment duration should be shortened to make solarization a practical technique.[8]

Biosolarization is a modified form of solarization that has been developed to address the duration, climate, and depth of efficacy issues associated with solarization.[12] Biosolarization couples soil microbial activity, which is induced through various soil amendments, with passive solar heating to generate multiple pest inactivation mechanisms in the soil (see Figure 1 for a general illustration of biosolarization plot establishment and weed inactivation effects). It can be thought of as combining elements of solarization and anaerobic soil disinfestation. These additional pesticidal mechanisms can include biological heating, soil oxygen depletion, and biological acidification of the soil via volatile fatty acid (VFA)
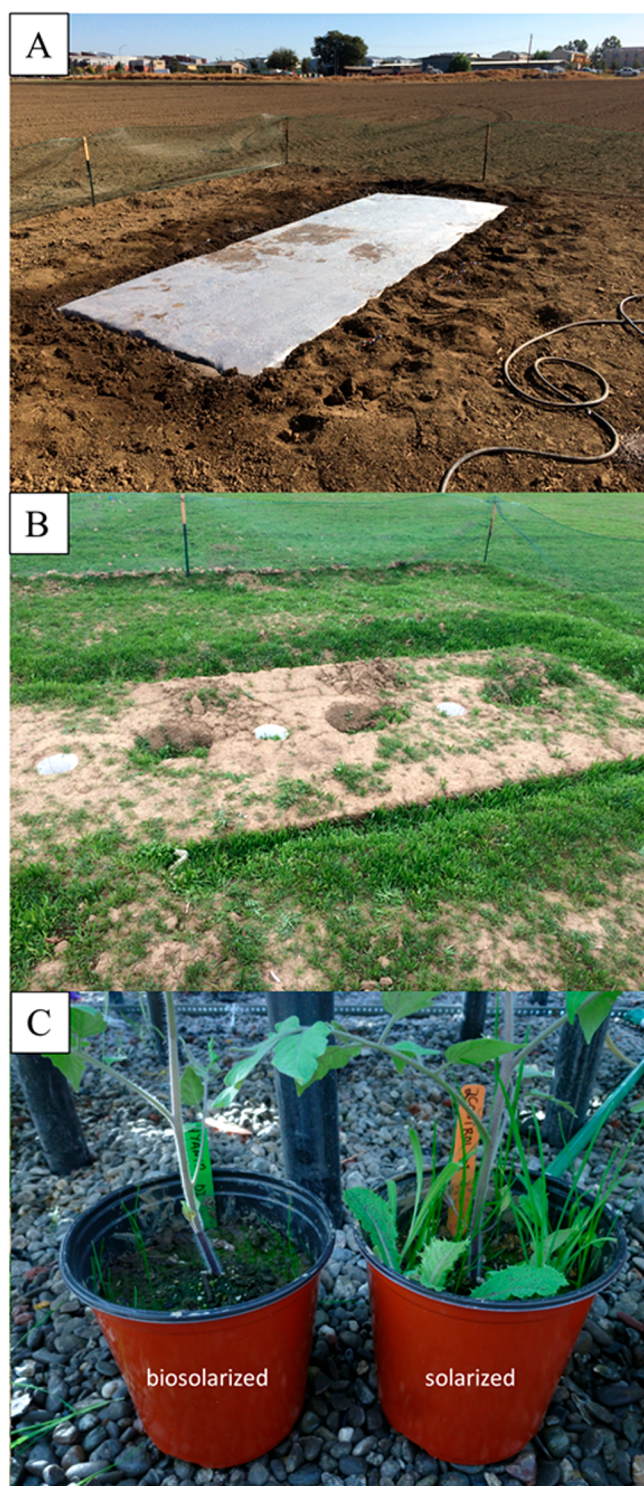
Figure 1. (A) A soil plot undergoing biosolarization. (B) A solarized plot 4 months after tarp removal. (C) Samples of solarized and biosolarized soils used for tomato cultivation in pots. The pots show the difference in residual viable weeds following treatment.

production.[13] Biosolarization has been shown to improve strawberry and tomato yields[14−16] as well as enhance soil fungal pathogen and nematode suppression in fruit and vegetable cropping.[17−19] Moreover, biosolarization can decrease the treatment duration to as little as 8 days and yield pest inactivation even when solar heating is sublethal[20,21].[13] Biosolarization can promote a circular economy by utilizing solid organic plant wastes

as soil amendments for agriculture.[22] Prior work has shown that mature green waste compost and wastes from industrial tomato (*Lycopersicum esculentum* L.) processing are effective biosolarization soil amendments.[13] Furthermore, expanded use of biosolarization in California would leverage California's Mediterranean climate and avoid or limit the need for post-planting weed and disease control measures.[23]

The objective of the present study was to compare solarization and biosolarization in terms of the generation of multiple potential pesticidal conditions and whether these changes to the soil environment affect the subsequent cultivation of tomatoes in the treated soil, with a focus on connecting initial phytotoxic conditions to agronomical risks and benefits for crop growth. To this end, the study aimed to determine the impact of biosolarization on the soil chemical environment during and immediately after treatment relative to that of solarization, as indicated by oxygen depletion and volatile fatty acid persistence at multiple depths. Agronomically and economically relevant biosolarization conditions were targeted, such as the use of a minimal treatment duration and the application of waste biomass as a soil amendment. To the authors' knowledge, this is the first study to measure a multitude of soil pest inactivation mechanisms, pest reduction, and tomato crop performance data in parallel for solarized and biosolarized soils. The findings will assist the commercial implementation of biosolarization using mature compost and industrial tomato waste amendments as a fumigation and herbicide alternative that can promote a circular economy via waste biomass recycling.

MATERIALS AND METHODS

Field Trial Experimental Framework. Two field experiments were conducted to investigate generation of pest-inactivating soil conditions during biosolarization and tomato growth in biosolarized and solarized soils. Both trials used mesocosms as experimental units and employed the same field site, as detailed elsewhere. The first trial, performed in 2015, aimed to quantify soil heating, oxygen depletion, and VFA accumulation phenomena at multiple depths in the soil during biosolarization. Additionally, various physiological responses in tomato plants grown in treated soil, which span vegetative growth, photosynthesis, and yield, were studied.

The second trial, completed in 2016, replicated the treatments of the initial trial to explore temporal variability in biosolarization as related to the persistence of soil VFAs and tomato plant growth responses. Moreover, it expanded upon the plant and fruit analyses to include gradation of fruit and chlorophyll content. A summary of the responses considered in each field trial is provided in Table 1. Descriptions of the methods used to measure each response are provided in subsequent sections.

Mesocosm and Field Site Preparation. Soil mixtures for mesocosms were prepared using soil collected from the field site. The soil was a Yolo silty clay loam (21% sand, 51% silt, and 28% clay) with 6% organic matter. To prepare the amended soil for the field trial, dry topsoil was collected from the upper 0−15 cm of the field site. The soil was sieved through a 3.18 mm screen to homogenize the samples and remove large particles. Soil amendments were obtained and processed as described elsewhere.[13] Briefly, tomato pomace (TP), the waste skins and seeds from industrial tomato paste production, was collected from a commercial processing facility during the 2014 harvest season. Mature green waste compost (GWC) generated from yard clippings was obtained from a commercial composting site in Zamora, CA in 2015. The composting facility monitored metal and pathogen levels to ensure the compost was suitable for soil application per US Environmental Protection Agency 40 CFR 503.13 and 40 CFR 503.32. Previous work has shown that this particular compost is mature and does not induce significant microbial activity when added to the soil as the sole amendment.[24] Rather, the compost is

Table 1. Responses Measured Across Both Field Trial Years

| responses	2015 field trial	2016 field trial
potential pest inactivation mechanisms in soil		
oxygen level	measured at 5 and 15 cm depth	not measured
pH	measured for well-mixed soil sampled from three depth levels: 0−7.5 cm, 7.5−15 cm, and 15−22.5 cm	not measured
temperature	measured at 0, 7.5, and 15 cm depth	measured at 5 and 15 cm depth
volatile fatty acid level	measured for well-mixed soil sampled from three depth levels: 0−7.5 cm, 7.5−15 cm, and 15−22.5 cm	measured for well-mixed soil collected from 0−22.5 cm depth
weed inactivation	measured in treated soil 6 months after treatment	measured in treated soil 15 days after treatment
tomato cultivation		
°Brix	measured	measured
average single fruit weight	measured based on all fruit	measured separately for ripe and unripe fruit
chlorophyll content	not measured	measured
fruit number	measured for all fruit	measured with delineation between ripe and unripe fruit
harvest index	measured	measured
photosynthesis	measured	measured
seedling germination	measured	not measured
stomatal conductance	measured	measured
total fruit weight	measured based on all fruit	measured separately for ripe and unripe fruit
vegetation fresh weight	measured	measured

destabilized by the addition of labile tomato pomace to enhance accumulation of biopesticidal organic acids in the soil.[25] Amendments were sun-dried after collection and then stored under ambient conditions. To improve amendment uniformity, dried TP was processed in a laboratory blender to reduce the particle size to less than 1 mm. The water holding capacity, pH, ash content, and bulk density of the amendments have been characterized previously.[24] Soil was amended to achieve 2.5% TP and 2% GWC (dry weight basis) for all the biosolarized treatments, as this combination of amendments has been shown to induce pesticidal soil conditions during biosolarization.[13] Non-amended soil was used for solarized treatments.

Mesocosms containing amended or non-amended soil served as experimental units for the field trial.[13] Mesocosms were constructed by loading soil into 3.8 L plastic soil growth bags (New England Hydroponics, Southampton, MA) that contained drainage holes to facilitate moisture and gas exchange with the surrounding soil in the field. When filled, the mesocosms measured 15 cm in diameter and 22.5 cm in height. In the 2015 field trial, as mesocosms were filled, miniature temperature sensors and data loggers (Thermochron iButtons model 1922L, Embedded Data Systems, Lawrenceburg, KY) were placed in a subset of mesocosms at 15, 7.5, and 0 cm (surface) depths. In the 2016 trial, temperature loggers were embedded at 5 and 15 cm depths. Furthermore, in the 2015 trial, oxygen sampling systems were embedded at 5 and 15 cm depths in the same subset of mesocosms to allow for sampling of the soil atmosphere while mesocosms were undergoing either solarization or biosolarization (Figure 2A). Each oxygen sampling system was comprised of a roughly 36 cm length of 3.18 mm porous soaker hose that was fashioned into a loop and embedded in the soil within each mesocosm. Soaker hoses were attached to a 3.18 mm tee to connect them to blunt needles (23 gage) via a Luer fitting, which in turn connected to a 122 cm length of polyethylene tubing (PE-50, inner diameter = 0.58 mm, outer diameter = 0.97 mm). The tubing was similarly attached to a valve via a needle and Luer fitting. The valve acted as a sample port and a controlled access to the soaker tube lumen. The sampling port valve was attached to a 3-way Luer valve, which controlled access to two syringes: a 3 mL syringe used to purge the line, fittings, and valves with soil gas from the embedded soaker hose and a 1 mL syringe subsequently used to extract a sample

of soil gas for further analysis. The volume within the polyethylene tubing, fittings, and valves was ≤2 mL, representing the minimum volume to be purged from the system ahead of sampling. Once assembled, the mesocosms were placed in a bath containing an excess of distilled water to permit wetting to field capacity by capillary action. Mesocosms were sealed in plastic tubs and incubated at 4 °C overnight to allow for moisture equilibration.

The field site was located at the Joe A. Heidrick, Sr., Western Center for Agricultural Equipment in Davis, CA (38.5°N; 121.8°W; elevation 16 m a.s.l.). The field was used as a teaching site for agricultural machinery operation for five years prior to the 2015 field trial and in the interim period between the 2015 and 2016 trials. During each of these years, the site was used to demonstrate discing, tilling, and other tractor-based operations from mid-April to July. The site was otherwise left undisturbed. The site was prepared as previously described.[13] Briefly, the site was plowed in two directions with a disc harrow to loosen and mix the topsoil and eliminate actively growing weeds, then smoothed with an orchard float. The site was irrigated 7, 5, 3, and 2 days before the field experiment using garden sprinklers that irrigated the entire field site. An additional irrigation was performed immediately before covering the soil with transparent plastic film. In total, approximately 6.5 cm of water was applied the week before the experiment. This was sufficient to bring the soil moisture content to field capacity ($\sim$27% (g_{water}/g_{soil})) at the depths sampled in this study.

The field site was separated into five replicate plots. Each plot measured 1.8 by 5.5 m. The plots were arranged in a row with a 1.8 m buffer zone between each of them. The mesocosms were buried in the field as previously described[13] (Figure 2B), and where applicable, the tubing for the soil atmosphere samplers was positioned such that attached sampling ports laid outside of the plot boundary. In the 2015 trial, each plot contained four mesocosms, representing duplicates of each amendment treatment: TP- and GWC-amended soil for biosolarization and non-amended soil for solarization. As described in subsequent sections, one duplicate was used for measuring soil temperature, oxygen content, moisture content, pH, VFA levels, volatile solids, and tomato growth parameters during and/or after treatment, while the other duplicate was used to measure weed survival and
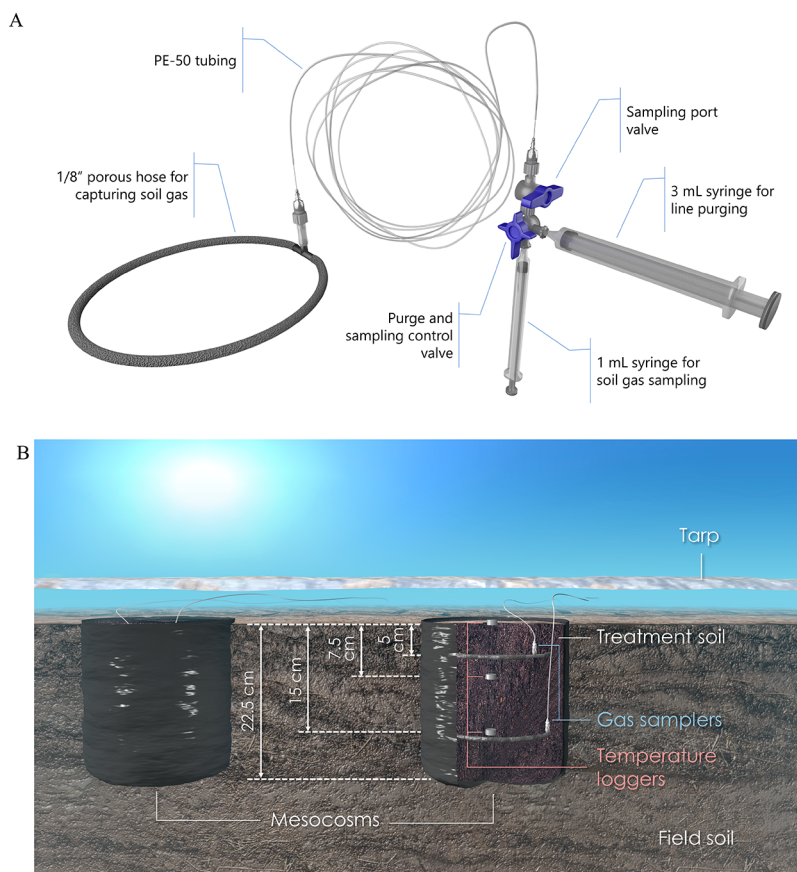
Figure 2. Mesocosm preparation and orientation in the field. (A) Certain mesocosms contained an apparatus to sample the soil atmosphere. Each apparatus consisted of a porous hose embedded in the soil to collect soil gas, which was connected via a length of tubing to a sample port located outside of the tarped plot. Syringes connected to the sample port were alternately used to purge ambient air from the tubing and fittings with soil gas and to extract a soil gas sample. (B) The illustration depicts a cross-section of soil containing two mesocosms as they would appear in the field during biosolarization. The mesocosm on the left shows a cutaway to reveal the orientation of the contents. Mesocosms used for soil temperature and gas monitoring contained two soil atmosphere samplers and three temperature loggers positioned at various depths.

growth 6 months after the treatment. In the 2016 trial, the plot layout was the same as that used in the 2015 trial except only two mesocosms, one for each biosolarized and solarized treatment, were placed in each plot, and only soil temperature, VFA level, and tomato growth data were obtained. The experimental site was surrounded by a chicken wire fence to prevent any intrusion by local fauna.

Field Experiment. Soil treatment was initiated on September 9, 2015 for the first field trial and on July 14, 2016 for the second field trial. Treatment initiation involved covering each freshly irrigated plot with 0.7 mil, transparent, low density polyethylene film (Huskey Film Sheeting; Poly-America, Inc., Grand Prairie, TX) and burying sheet edges in the soil along plot borders. Care was taken to minimize the headspace beneath the film. In the 2015 trial, the soil atmosphere was sampled periodically during the experiment. This was accomplished by opening the sampling port, flushing the sampling system with 3 mL of soil gas, followed by collection of a 1 mL sample of soil gas. The first oxygen sample had a technical problem and was not included as a valid sample, but from the second day of the experiment until the sixth day, evening (17:00−19:00) and morning (05:00−07:00) samples were taken.

Each field experiment lasted 8 days, after which the plastic film was removed from the field site. The mesocosms designated for tomato growth studies were left in the field to aerate for an additional 12 days in the 2015 trial or 15 days in the 2016 trial to allow for remediation of any residual phytotoxic conditions. In the 2015 trial, the mesocosms allocated for measurement of weed inactivation were left in the field for six months after tarp removal. In the 2015 trial, exhumed mesocosms were sectioned into three 7.5 cm-thick layers for subsequent analysis of various properties by soil depth, as described in the

following sections. In the 2016 trial, the mesocosms were not sectioned and VFA levels were measured for the bulk mixture.

Soil Characterization. Soil pH, volatile solids, moisture content, VFA levels, and oxygen content were measured at various depths for mesocosms in the 2015 trial, whereas only VFA levels were examined in the 2016 trial mesocosms. The moisture content, pH, and VFA measurements are described in depth elsewhere.[13] Briefly, moisture content was measured gravimetrically by comparing soil mass prior to and following drying in an oven at 105 °C. Soil samples were extracted for VFA and pH measurements by combining soil with water at a 1:1 mass ratio. Formic, acetic, propionic, isobutyric, and butyric acid contents in the extracts were analyzed by HPLC instrumentation (model UFLC-10Ai, Shimadzu, Columbia, Maryland, USA) using an Aminex HPX-87H column (300 × 7.8 mm) (Life Science Research, Education, Process Separations, Food Science, Hercules, CA, USA) and an SPD-M20A photodiode array detector set at 210 nm (Shimadzu). Extracts were run as previously described.[25] Measured VFA concentrations were normalized according to the moisture content of each extracted sample to yield concentration per unit dry weight of soil, and the total VFA content was calculated by summing the entire VFAs measured in each sample. Volatile solids content was determined based on the mass of the soil samples following incineration at 550 °C for 7 h.

Soil atmosphere samples were immediately characterized after collection using GC/TDC instrumentation (Agilent 6890N with a Hayesep DB 100/120 column 40′ × 1/8″ × 0.085″ SS) equipped with a thermal conductivity detector (Agilent). The GC program started with a 10 min hold at 30 °C, ramp at 20 °C/min to 50 °C with a hold for 8 min, ramp at 20 °C/min to 70 °C with a hold for 12 min, and a ramp at 30 °C/min to 100 °C with a hold for 1 min. The flow rate

through the column was 45 mL/min. An ambient air sample was used as a standard to identify baseline nitrogen and oxygen peaks in the chromatograph. The oxygen peak was used as an internal standard for the samples from the field. A standard curve was also made for varying amounts of oxygen in the range of 21−2.1%.

Greenhouse Tomato Growth Study. In the 2015 trial, after removal of samples from each mesocosm depth section (approximately 50 g) for the previously described soil characterization, soils from all biosolarized and solarized mesocosms were pooled according to treatment to create two stock mixtures of soil. Each stock of biosolarized and solarized soil was well-mixed and used for subsequent tomato cultivation tests. Likewise, well-mixed samples of soil spanning the entire depth of each mesocosm were used in the 2016 trial. All growth experiments were conducted in a greenhouse maintained between 18 and 28 °C and 50−70% relative humidity.

Germination Assay. Tomato germination assays were done using soil samples from the 2015 trial. Seeds of processing tomatoes (cv. SUN6366, Nunhems USA, Inc., Parma, ID) were germinated in biosolarized and solarized soils to gauge residual phytotoxicity. The viability of the seed lot was 95% based on vendor specifications. Each experimental unit consisted of 10 seeds sown at approximately 1 cm depth in 2.37 L pots (SP-630, East Jordan Plastics). Pots were fertigated twice daily with 300 mL of water containing 143 mg/L N (delivered as 136 mg/L NO_3-N and 7 mg/L NH_4-N), 63 mg/L P (delivered as $H_2PO_4^-$), 199 mg/L K^+, 125 mg/L Ca^{2+}, 49 mg/L Mg^{2+}, 65 mg/L S (delivered as SO_4^{-2}), 2 mg/L Fe^{3+}, 0.097 mg/L Cu^{2+}, 0.633 mg/L Mn^{2+}, 0.055 mg/L Mo^{6+}, and 0.097 mg/L Zn^{2+}. After 2 weeks, emerged seedlings were counted to determine the germination percentage. Seedlings were dried for 24 h at 105 °C and then weighed to determine dry biomass.

Tomato Growth. To simulate commercial processing tomato cultivation, where greenhouse grown plants are later transplanted to the field for maturation and fruit production,[26] tomato seeds were germinated under greenhouse conditions and then transferred to pots containing biosolarized or solarized soil for the remainder of the growing period. Specifically, tomato seeds were germinated in germination trays in a commercial potting soil mixture (Hastie's Capitol Sand and Gravel; 25% screened topsoil, 5% lava fines and sand, and 70% mixture of equal parts forest humus, composted fir, and compost from horse manure and wheat straw). Approximately 2 weeks after germination, seedlings of approximately the same size were transplanted into pots containing the field soils. To prepare the pots, 2.37 L pots were filled with equal amounts (around 2 L of soil per pot) of biosolarized or solarized soil. Six replicate pots in 2015 and five in 2016 were used for biosolarized and solarized soils (corresponding to the maximum number of pots that could be filled with the soil available). Soils were initially saturated with the previously described fertilizer water. Plants were fertigated daily with 400 mL of solution. All pots were hand weeded after 3 weeks of growth. Plants and fruit were harvested after 3.5 months, and various morphological measurements were made as described in the following sections.

Photosynthesis, Gas Exchange, and Chlorophyll Content Measurements. For both 2015 and 2016 trials, measurements of photosynthesis and stomatal conductance were made on fully expanded leaves of plants after 4 weeks of growth. A Li-6400 portable gas-exchange system (LI-COR) was used to collect gas exchange data related to photosynthesis and stomatal conductance. Photosynthesis was induced by saturating an enclosed section of leaf with light (1,000 μmol m^{-2} s^{-1}) in the presence of 400 μmol mol^{-1} CO_2 surrounding the leaf. The amount of blue light was set to 10% photosynthetically active photon flux density to optimize stomatal aperture. The temperature was set to 25 °C. In the 2016 trial, additional measurements of leaf chlorophyll were obtained. For chlorophyll extraction, the leaves were weighed and ground in liquid N_2 and then crude lysates were extracted in 80% acetone. The absorbance at 663 and 645 nm was measured in the extracts using a spectrophotometer (DU-640; Beckman Coulter). Total chlorophyll content for each sample was calculated as described elsewhere.[27]

Tomato Plant Analyses. At harvest, the above-ground biomass of the plants was cut, the fruit were picked and counted, and the weight of the above-ground vegetation was measured. The total combined fresh weight of fruit was measured for each plant. The average single fruit weight was calculated by dividing total fruit weight for a given plant by the number of fruit present. In the 2016 trial, the harvested fruit were divided into two grades: fully ripe, red tomatoes (grade 1) and unripe green tomatoes (grade 2). The harvest index was calculated as the quotient of the total fruit weight divided by the sum of the vegetation fresh weight and the total fruit weight. The sugar concentration in a representative subset of mature ripe fruit (°Brix) was determined using a digital refractometer (PR-100, ATAGO USA, Inc., Bellevue, WA).

Weed Seed Inactivation. Emergent weed plants at the field site included cool season monocots (Poaceae), broadleaf thistles (Asteraceae), mustards (Brassicaceae), and fiddleneck (Boraginaceae) during the soil sampling period of the 2015 field trial. In the 2016 trial, warm season monocots (Poaceae, Cyperaceae), broadleaf thistles (Asteraceae), amaranths (Amaranthaceae), morning glory (Convolvulaceae), and caltrop (Zygophyllaceae) were the emergent weeds during soil sampling. These weeds are common to agricultural settings in California's central valley.[28] The endogenous distribution of seeds from these plants formed the basis for assessing weed seed inactivation following solarization or biosolarization.

In the 2015 trial, five mesocosms from each treatment were left in the field for 6 months after tarp removal to allow for complete dissipation of any induced seed dormancy factor(s). Mesocosms were exhumed and processed as described previously. Three replicate 1 L pots were filled with equal amounts (around 2/3 of the pot's volume) of biosolarized or solarized soil from each of the mesocosm soil depth sections examined. Pots were then saturated with the previously described fertigation solution and incubated in the greenhouse for 3 weeks to allow for germination and growth of any viable weed propagules that remained in the soil. Emergent weed plants were then counted, carefully harvested, dried at 105 °C overnight, and then weighed to determine dry biomass. In the 2016 trial, weed seed inactivation was determined immediately following solarization or biosolarization, i.e., the combined effect of mortality and dormancy, to complement the 2015 data that aimed to measure seed mortality separate from dormancy. To this end, weed biomass was harvested from pots used in the 2016 tomato growth study 3 weeks after tomato plants were transplanted into the pots. The fresh weight of the harvested weed biomass was measured for each pot.

Data Analysis. Comparison of response means between treatments was conducted via one-way ANOVA with post-hoc Tukey's Honest Significant Difference test. A family-wise error rate of 0.05 was used for every comparison. Statistical analyses were performed using JMP Pro software (version 12.0.0, SAS, Cary, NC).

■ RESULTS

Soil Heating. At all depths examined, biosolarization and solarization resulted in similar soil temperatures across both field trials (Figure 3). Although the solarization procedure in 2015 was carried out rather late in the season (September), the temperature was as expected in soil solarization and reached a maximum of 50 °C near the surface (Figure 3A). In 2015, the difference between the daily peak solarized surface temperature and the ambient temperature was generally 10−20 °C (Figure 3A). For both trial years and all depths examined, temperature profiles were similar for solarized and biosolarized soils (Figures 3A−E). The 2016 trial compared the soil temperature in treated plots to that of untreated soil at the 15 cm depth. At this depth, daily peak temperatures were similar between the treated and untreated soils, but the daily minimum temperature remained 5−10 degrees greater in the treated plots compared to the untreated soil.

Soil Physical and Chemical Properties. The impact of biosolarization with TP and GWC on various soil properties was measured in the 2015 field trial (Table 2, Figure 4). As expected, the incorporation of TP increased the soil organic
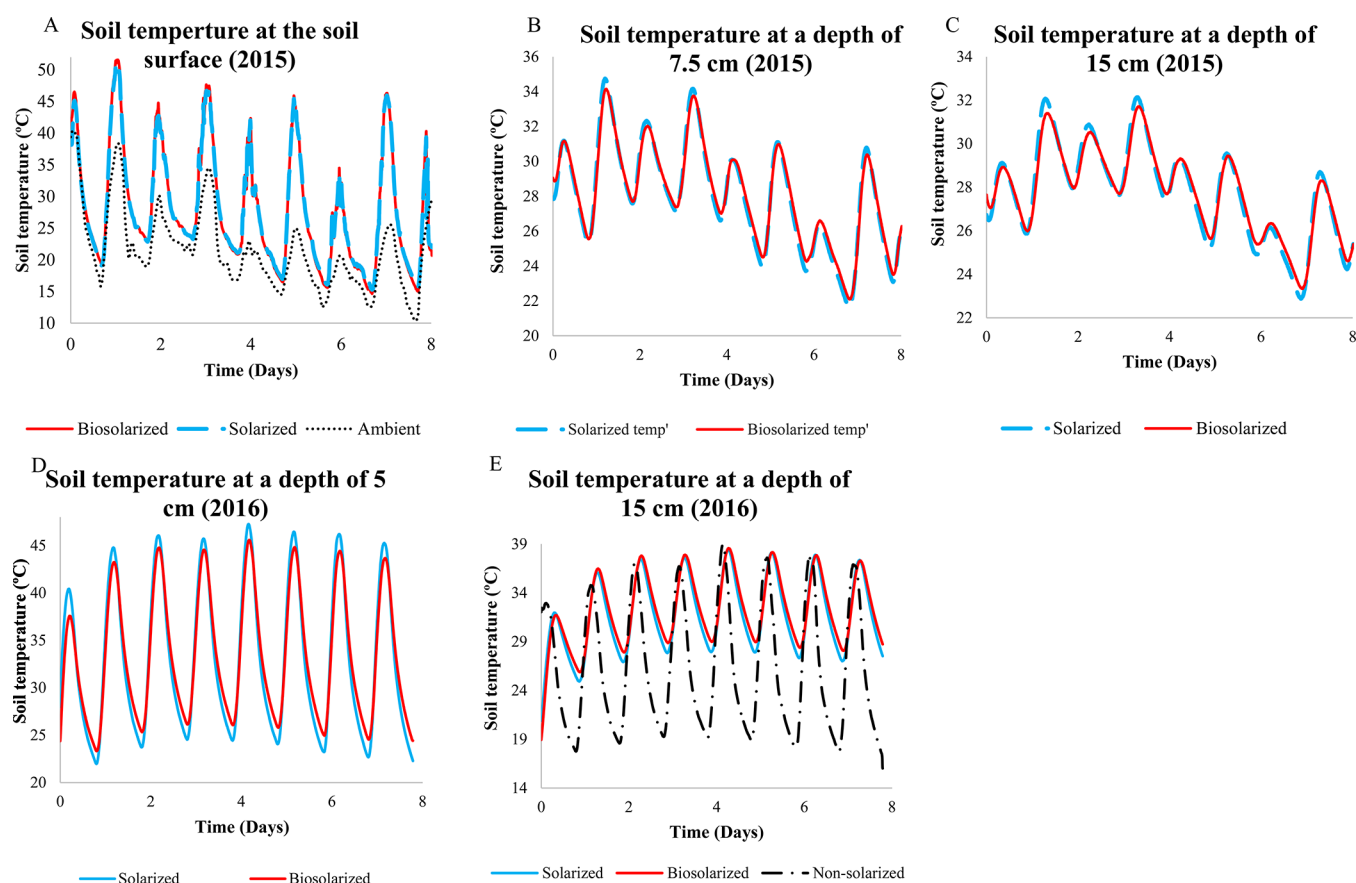
Figure 3. Soil temperature time courses at various depths in solarized and biosolarized mesocosms during the 8 day treatment period. Temperature data are presented for 2015 at a depth of (A) 0, (B) 5, and (C) 15 cm and 2016 at a depth of (D) 5 and (E) 15 cm. The dotted line (A) in the 2015 trial represents the ambient temperature measured at a nearby meteorological station, and the dash-dotted line in (E) indicates the temperature in non-solarized control soil at a 15 cm depth.

Table 2. Properties of Solarized and Biosolarized Soil Samples Taken from Field Mesocosms[a]

| soil treatment	depth (cm)	moisture (g_{water}/g_{soil})[b]	VS (% DS)[b]	pH[b]	VFAs (mg/g_{DW})[b,c]
solarized soil sampled from field plots (2015 trial)	0−7.5	0.22 ± 0.01 (b)	6.12 ± 0.62 (b)	7.03 ± 0.25 (a)	N.D.
	7.5−15	0.22 ± 0.01 (b)	6.21 ± 0.47 (b)	7.28 ± 0.37 (a)	N.D.
	15−22.5	0.23 ± 0.01 (b)	6.12 ± 0.42 (b)	7.33 ± 0.32 (a)	N.D.
biosolarized soil sampled from field plots (2015 trial)	0−7.5	0.26 ± 0.01 (a)	7.87 ± 0.80 (a)	7.71 ± 0.37 (a)	0.01 (b)
	7.5−15	0.27 ± 0.02 (a)	8.17 ± 0.69 (a)	7.56 ± 0.20 (a)	0.59 (b)
	15−22.5	0.28 ± 0.01 (a)	8.09 ± 0.89 (a)	7.23 ± 0.58 (a)	2.86 (a)
solarized soil after use for greenhouse tomato growth (2015 trial)	−	0.22 ± 0.02 (A)	6.58 ± 0.16 (C)	6.97 ± 0.17 (A)	N.D.
biosolarized soil after use for greenhouse tomato growth (2015 trial)	−	0.24 ± 0.02 (A)	7.95 ± 0.30 (B)	7.24 ± 0.35 (A)	N.D.
solarized and biosolarized soil sampled from field plots (2016 trial)	−	−	−	−	N.D.

[a](−), not measured; VS, volatile solids; DS, dry solids; and VFA, volatile fatty acids. [b]A comparison of response means between treatments was conducted via one-way ANOVA with post-hoc Tukey's Honest Significant Difference test. Values that do not share a letter are significantly different ($P < 0.05$). Within each column, lowercase letters represent comparisons among values measured after the 12 day remediation period following tarp removal from the field. Uppercase letters represent comparisons between values measured after three months of incubation in the greenhouse. [c]Estimated value based on the sum of measured values for formic, acetic, propionic, isobutyric, and butyric acids. N.D., not detected.

matter content. Prior to treatment in the field the volatile solids contents for amended and non-amended soils were 9.49 ± 0.42 and 6.16 ± 0.26% (w/w), respectively. The biosolarization treatment reduced the volatile solids content of the amended soil to 7.87 ± 0.80% (w/w) due to microbial consumption (Table 2), while solarization did not affect the volatile solids content of the non-amended soil. The increased residual organic matter in the biosolarized soil translated to an increased soil water holding capacity. At the conclusion of the field

experiment, the biosolarized soil retained 0.04−0.05 g_{water}/g_{soil} more than the non-amended solarized soil (Table 2). The pH of the amended soils was similar to those of the non-amended soils 2 weeks after tarp removal and was approximately neutral (Table 2).

VFAs were detected only in the biosolarized mesocosms from the 2015 trial. The residual VFA content increased with greater depth in biosolarized soil and reached a maximal of 2.86 $mg/g_{dry\ soil}$ in the lower depths of 15−22.5 cm.
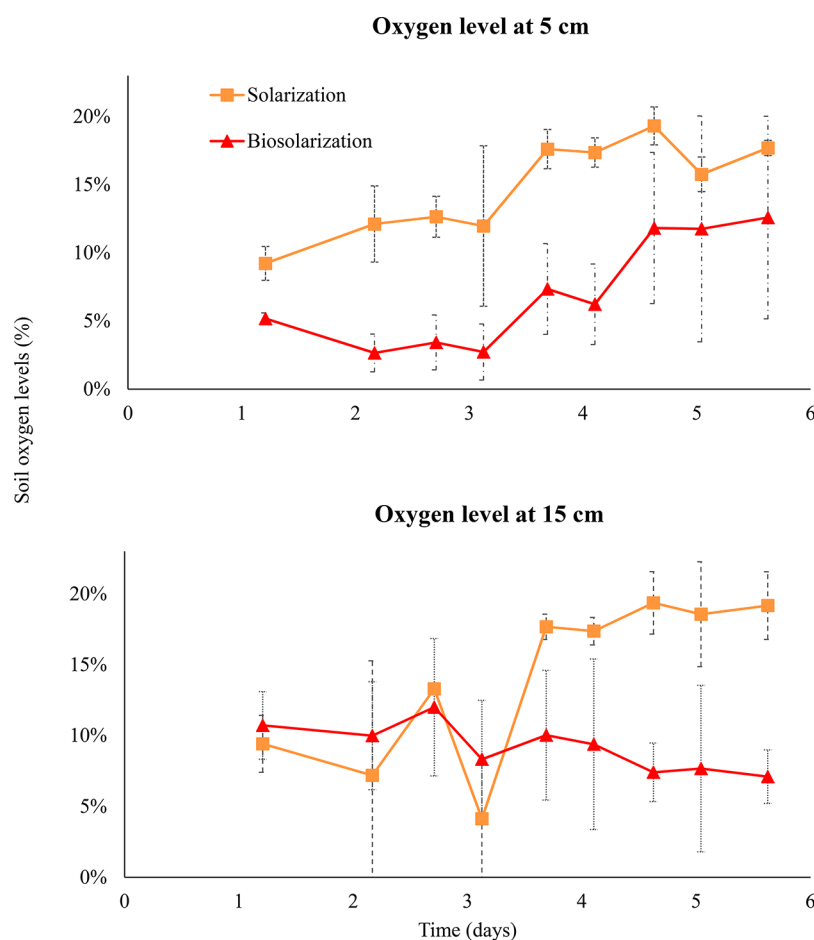
Figure 4. Soil oxygen levels in the 2015 trial at 7.5 and 15 cm depth in the solarized and biosolarized mesocosms. The data points represent three replicate measurements. The replicates were taken in the morning and evening of each sampling day. Error bars indicate one standard deviation.

The quantity of residual VFAs in the deepest soil layer did not result in acidic pH values (Table 2). After transfer to pots for greenhouse cultivation of the tomatoes, no residual VFAs were detected in the soil at the time of tomato harvest. In 2016, no VFAs were detected across all of the treatments.

The oxygen level data (Figure 4) showed that both solarization and biosolarization led to soil oxygen depletion during treatment (Figure 4). In the upper layer (0−5 cm), biosolarization resulted in greater decreases in oxygen (achieving concentrations between 3% and 12% oxygen) than solarization (9.5−18% oxygen) throughout the 8 day treatment (Figure 4). At greater soil depths, the oxygen concentration trend was different, and both the solarized and the biosolarized soils were anaerobic or microaerobic (8−10% soil oxygen). Three days after initiating the treatment, oxygen levels in solarized soils were close to that of ambient air (18−20% soil oxygen) while in the biosolarized mesocosms levels continued to decrease (7−9% soil oxygen) (Figure 4).

Weed Inactivation. The 2015 field trial investigated weed seed survival several months after soil treatment. In the six months following solarization and biosolarization, weeds germinated and grew in the areas surrounding the experimental plots but fewer grew in the tarped areas of the plots and none grew within the mesocosms that remained in the field (Figure 1B). To discriminate between weed seed dormancy and seed death at 6 months post-treatment, mesocosm soil samples were potted, wetted, and incubated under ideal growth conditions in a greenhouse to break any dormancy and promote germination.

Weed germination and growth were measured for each soil treatment and depth (Table 3). For solarized soil, the uppermost soil layer (0−7.5 cm) exhibited the greatest seed mortality compared to the deeper layers (5.8 ± 3.11 weeds/pot in the 0−7.5 cm depth vs 14.4 ± 6.35 and 9.4 ± 1.52 at 7.5−15 and 15−22.5 cm depths, respectively). Biosolarized soils showed 1.8 ± 1.64 weeds/pot in the uppermost soil layer and complete seed mortality in the lower two layers, which was significantly greater than the inactivation that was achieved by solarization at the two lower depths. Weed biomass data followed similar trends (Table 3, Figure 1C).

The 2016 trial examined weed seed inactivation approximately 5 weeks after the conclusion of soil treatment. Significant differences in weed biomass were observed between the solarized and the biosolarized soils (Table 3). Whereas the biosolarized soil showed very low weed biomass (0.008 ± 0.017 kg/pot), solarized soil exhibited considerable weed growth (0.378 ± 0.357 kg/pot).

Tomato Plant Growth. Solarized and biosolarized soils from the field were used for tomato plant growth studies in two consecutive years (September 2015 and August 2016). Controlled greenhouse conditions were used to give optimal growth conditions and to avoid the substantial variability that field conditions can cause. Solarized and biosolarized soils yielded germination rates of 40 ± 18% and 20 ± 20%, respectively (in the 2015 experiment). This was substantially lower than the viability specified by the seed vendor (95%), indicating that various factors in the potted soil system, such as soil compaction, may

Table 3. Tomato Plant Germination Rate and Weed Suppression in Treated Soils[a]

soil treatment and preparation	depth (cm)	germination rate (%)	seedling dry biomass (g)	weed germination (no. of weed plants/pot)[b]	weed growth 6 months post-treatment ($g_{dry\ weight}$/pot)[b]	weed growth 15 days post-treatment ($g_{dry\ weight}$/pot)[c]
solarized soil sampled from field plots	0−7.5	−	−	5.8 ± 3.11 (b,c)	0.99 ± 0.72 (b,c)	−
	7.5−15	−	−	14.4 ± 6.35 (a)	2.36 ± 0.99 (a)	−
	15−22.5	−	−	9.4 ± 1.52 (a,b)	1.46 ± 0.544 (a,b)	−
biosolarized soil sampled from field plots	0−7.5	−	−	1.8 ± 1.64 (c)	0.35 ± 0.39 (c)	−
	7.5−15	−	−	0 (c)	0 (c)	−
	15−22.5	−	−	0 (c)	0 (c)	−
solarized soil transferred to greenhouse potted growth system	−	40 ± 18 (a)	0.01 ± 0.007 (a)	−	−	0.378 ± 0.357 (a)
biosolarized soil transferred to greenhouse potted growth system	−	20 ± 20 (a)	0.005 ± 0.003 (b)	−	−	0.008 ± 0.018 (b)

[a](−), value not measured. Values that do not share a letter are significantly different ($P = 0.05$). The comparison of response means between treatments was conducted via one-way ANOVA with post-hoc Tukey's Honest Significant Difference test. [b]Measured values for soil samples from mesocosms that remained embedded in the field for 6 months following treatment prior to greenhouse incubation. [c]Measured values for soil samples from the tomato plant pots after 15 days of soil remediation in the field and one month of growth in the greenhouse.

have affected germination across all treatments. Significantly lower seedling biomass was observed in biosolarized soil compared to that of solarized soil (Table 3), suggesting that inhibitory levels of VFAs or other phytotoxic compounds persisted in the soil for the 12 day remediation period following biosolarization. In light of this, the 2016 trial used a 15 day remediation period to promote greater dissipation of phytotoxins.

To mimic current cultivation practices, seeds were germinated and then healthy seedlings were transplanted into field soil. Various physiological properties were measured following plant growth in greenhouse-incubated pots containing biosolarized and solarized soils (Table 4). In the 2015 field trial, among several measured plant physiology and yield responses spanning photosynthesis, vegetative biomass, fruit weight and quantity, and tomato Brix value, there was no evidence that a significant difference existed between plants grown in solarized and biosolarized soils (Table 4).

Unlike the 2015 trial, ANOVA of the tomato cultivation data from the 2016 trial showed significant differences for several responses. Vegetation fresh weight, total fruit weight, and total fruit number were all significantly elevated in plants grown in the biosolarized soil compared to those grown in solarized soil (Table 4). Elevated total fruit mass stemmed from significant increases in both ripe and unripe fruit mass for plants grown in biosolarized soil. Furthermore, the number of fully ripe fruit was significantly higher for plants grown in biosolarized soil.

■ DISCUSSION

Former studies showed the potential for biosolarization to add additional heat to the soil via microbial activity to complement the solarization heating effect.[26,29] However, in this study there was no significant difference in soil temperature between the solarized and the non-solarized treatment. Similar temperature results, which are generally lower than those observed in other solarization work,[8,30,31] were also seen in a mesocosm field trial with the same amendments in a sandy loam soil and likely reflect cooler weather.[13,19] At all depths examined, soil temperature remained below 55 °C, the temperature typically targeted in composting to inactivate pathogens. Moreover, the data showed that temperatures in the 39 to 50 °C range, which are known to inactivate seeds from several major weed species,[32] were achieved down to 5 cm for a fraction of each day. However, temperatures fell below this range at lower depths. This highlights the need

for additional pesticidal factors to compensate for sublethal soil heating. The weed inactivation data agreed with this notion as solarized soils contained viable weed seeds after treatment while biosolarized soils largely did not.

In this study, soil oxygen levels are presented in relation to time and depth during solarization and biosolarization. To the authors' knowledge, this is the first time temporal and spatial soil oxygen data have been presented for these processes. The oxygen content profiles suggest that microbial consumption of oxygen may have occurred within the first 3 days of treatment for both solarization and biosolarization. However, solarized soils did not show a significant decrease in volatile solids content, indicating that perhaps organic carbon was immobilized in microbial biomass rather than metabolized to volatile products or was otherwise displaced or depleted through processes outside of fermentation. The additional oxygen depletion in the biosolarized soil over the first 6 days of treatment was likely due to microbial consumption of amended organic matter, as evidenced by the decrease in volatile solids.

Understanding oxygen depletion during biosolarization could unveil when anoxic stresses might affect pests and when biopesticidal anaerobic fermentation products may be produced. At both 5 and 15 cm depths, persistence of oxygen levels on the order of 10% or less was uniquely observed in the biosolarized soil over the 8 day treatment period. Studies of oxygen stress effects on phytoparasitic nematode mortality showed varying sensitivities. Phytoparasitic nematodes such as *Meloidogyne incognita*, *Trichodorus cristiei*, and *Tylenchus semipenetrans*, were affected only by oxygen levels <1% over a 10 day period.[33] Conversely, phytoparasitic *Xiphinema americanum* exhibited inactivation proportional to oxygen depletion (i.e., going from 21% to <1% oxygen) over incubation periods up to 10 days in length.[33] For desirable nematodes, such as the insect pest biocontrol agent *Steinernema carpocapsae*, 10% oxygen eliminated insect pathogenicity only after 4 weeks of exposure.[34] Together, these results suggest that the magnitude and duration of oxygen stress induced via biosolarization may be tuned to promote inactivation of certain phytoparasitic nematodes while preserving the activity of desirable entomopathogenic nematodes. However, more research is needed to explore this possibility and to gauge the effect of oxygen limitation on other soil pests. Moreover, there is a need to investigate interaction effects between oxygen supply and other

Table 4. Tomato Plant and Fruit Properties[a]

treatment	photosynthesis (μmol CO_2 m^{-2} s^{-1})	stomatal conductance (mol water m^{-2} s^{-1})	fruit number	fruit no. grade 1[b]	fruit no. grade 2[b]	total fruit weight (g)	fruit weight (g) grade 1[b]	fruit weight (g) grade 2[b]	vegetation fresh weight (g)	harvest index	average single fruit weight (g)	°Brix	chlorophyll (mg/gFW)
2015 Solarized	19.76 ± 2.24 (a)	0.28 ± 0.07 (a)	20.67 ± 6.02 (a)	N.D.	N.D.	342.58 ± 85.64 (a)	N.D.	N.D.	387.67 ± 65.48 (a)	0.47 ± 0.09(a)	17.39 ± 5.66 (a)	6.09 ± 0.58 (a)	N.D.
2015 Biosolarized	20.30 ± 1.39 (a)	0.30 ± 0.05 (a)	15.50 ± 7.15 (a)	N.D.	N.D.	330.92 ± 62.58 (a)	N.D.	N.D.	317.42 ± 66.21 (a)	0.51 ± 0.05(a)	24.01 ± 8.08 (a)	6.12 ± 0.58 (a)	N.D.
2016 Solarized	20.09 ± 6.87 (A)	0.35 ± 0.18 (A)	15.4 ± 9.61 (B)	8.4 ± 5.60 (B)	7.0 ± 5.48 (A)	418.00 ± 242.22 (B)	292.00 ± 201.67 (B)	126.00 ± 53.67 (B)	642.00 ± 213.70 (B)	0.37 ± 0.14 (A)	29.17 ± 8.49 (A)	6.24 ± 1.18 (A)	1.70 ± 0.19 (A)
2016 Biosolarized	21.55 ± 3.28 (A)	0.42 ± 0.06 (A)	36.6 ± 12.10 (A)	17 ± 2.55 (A)	19.6 ± 12.82 (A)	1240.00 ± 167.78 (A)	818.00 ± 60.17 (A)	422.00 ± 159.75 (A)	1092.00 ± 89.27 (A)	0.53 ± 0.03 (A)	36.46 ± 1.13 (A)	6.23 ± 0.68 (A)	1.65 ± 0.22 (A)

[a]Values are given as mean ± standard deviation. n = 6 for 2015; n = 5 for 2016. Within each column, values that do not share a letter are significantly different (P ≤ 0.05). Lowercase letters denote comparisons within the 2015 data, while uppercase letters refer to comparisons within the 2016 data. The comparison of response means between the two treatments was conducted by Student's paired t-test. N.D., not determined. [b]Fully ripe, red tomatoes (grade 1) and unripe green tomatoes (grade 2).

biosolarization stresses, such as elevated temperature and biopesticide accumulation, as they relate to pest mortality.

In light of the moderate soil heating, fermentation processes in the soil may have been the primary driver of biocidal conditions in the soil as they are with anaerobic soil disinfestation.[35] Such microbial activity during biosolarization can result in lower soil pH and VFA accumulation.[3,13,24,26] Previous studies have demonstrated that pH depression in biosolarized soils can lead to phytotoxic soil conditions.[13,24] If such acidification occurred in the present study, the pH reverted to its original value within the remediation period. Although there was no lasting effect on the pH, VFA production was detected in biosolarized soils. The importance of VFAs as biopesticides has been demonstrated.[26,36] In the current study, residual VFAs were primarily detected beyond a 15 cm depth in the 2015 trial where a shorter remediation period was used. This was likely due to the upper soil layers preventing diffusion of oxygen from the surface and inhibiting diffusion of VFAs to the surface. Both phenomena promote greater production and retention of VFAs in the soil.[24] The observed persistence of more oxygen-limited conditions at 15 cm depth relative to 5 cm depth is consistent with this notion. Similar oxygen depletion kinetics have previously been observed during biosolarization with wild garden rocket and thyme amendments.[37] Additional research is needed to elucidate any direct contribution of oxygen depletion to weed seed death outside of fostering anaerobic fermentation and VFA production.

Biosolarization is geared toward creating phytotoxic soil conditions for weed inactivation. However, the persistence of those conditions post-treatment poses risks for subsequent crops, particularly when short biosolarization and remediation periods are used and phytotoxic compounds, such as VFAs, are given less time to dissipate from the soil. While prior work has used similar compost and tomato waste amendments for biosolarization, it did not look beyond VFA generation and disinfestation to investigate crop production.[25] For the first time, this study compared residual phytotoxicity following a brief treatment (8 days) and remediation period (12 to 15 days) for solarization and biosolarization with respect to a wide array of growth and metabolic characteristics for tomatoes. The results indicated that 12−15 days of remediation after removal of the plastic tarp could be sufficient to largely eradicate any residual phytotoxicity. The two field trials agreed that any lingering phytotoxicity from biosolarization did not negatively impact plant health with respect to plants grown in solarized soil. Whereas the 2015 trial did not yield evidence of significant differences between plants grown in biosolarized and solarized soils, the 2016 trial showed significant improvement in plant performance with elevated vegetative biomass, total fruit quantity, and number of ripe fruit. It is notable that improved plant performance was seen only in the year where no residual VFAs were detected in the soil at the time of planting. Phytotoxicity from VFAs has long been documented in anaerobic soils amended with organic materials.[38] A previous laboratory study showed that phytotoxicity associated with a GWC amendment was ameliorated when the amended soil was subjected to soil heating.[29] Furthermore, the residual phytotoxicity is likely linked to the short treatment duration and the brief 12 day remediation used in the 2015 trial. Longer biosolarization times or remediation periods after tarp removal may allow VFAs and other phytotoxins to dissipate completely ahead of crop growth and allow for improved plant growth in biosolarized soils, as observed in the 2016 trial. The 2016 field trial data indicate that a remediation period as short as 15 days

may be suitable. However, additional research is needed to determine how environmental factors such as soil texture, temperature, rainfall, wind speed, and microbiological activity may affect the rate of phytotoxin dissipation. Furthermore, research is needed to understand the mechanisms behind the improved plant performance seen in the 2016 trial. Such mechanisms could include decreased competition from weeds, suppression of pathogens and phytoparasites, and benefits from the addition of organic matter to the soil during biosolarization, such as improved soil structure and stability, cation exchange capacity, phytonutrient levels, and beneficial microorganism content. Beyond factors that directly affect crop growth, research is also necessary to understand broader changes to soil biodiversity following solarization or biosolarization that could impact the local food web or responses to environmental perturbations.

In conclusion, the data indicate that biosolarization may be a more effective technique for weed control compared to solarization. Biosolarization is capable of inactivating weed seeds with a short treatment duration and can achieve inactivation at greater depths where solarization alone is ineffective. However, the prospect of greater weed inactivation must be balanced against the potential for lingering phytotoxicity in the soil following biosolarization. Such phytotoxicity is likely linked to residual VFAs in the soil. Altering the type and concentration of soil amendments may prevent excessive accumulation of phytotoxins during biosolarization. Furthermore, adjusting the treatment duration or length of the remediation period following biosolarization may allow phytotoxins to dissipate ahead of crop cultivation. Additional research is needed in these areas to optimize biosolarization for compatibility with tomatoes and other target crops.

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