

The Effects of Anaerobic Soil Disinfestation on Weed and Nematode Control, Fruit Yield, and Quality of Florida Fresh-market Tomato

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Abstract. Anaerobic soil disinfestation (ASD) is considered a promising sustainable alternative to chemical soil fumigation (CSF), and has been shown to be effective against soilborne diseases, plant-parasitic nematodes, and weeds in several crop production systems. Nevertheless, limited information is available on the effects of ASD on crop yield and quality. Therefore, a field study was conducted on fresh-market tomato (*Solanum lycopersicum* L.) in two different locations in Florida (Immokalee and Citra), to evaluate and compare the ASD and CSF performances on weed and nematodes control, and on fruit yield and quality. In Immokalee, Pic-Clor 60 (1,3-dichloropropene + chloropicrin) was used as the CSF, whereas in Citra, the CSF was Paldin™ [dimethyl disulfide (DMDS) + chloropicrin]. Anaerobic soil disinfestation treatments were applied using a mix of composted poultry litter (CPL) at the rate of 22 Mg·ha⁻¹, and two rates of molasses [13.9 (ASD1) and 27.7 m³·ha⁻¹ (ASD2)] as a carbon (C) source. In both locations, soil subjected to ASD reached highly anaerobic conditions, and cumulative soil anaerobiosis was 167% and 116% higher in ASD2 plots than in ASD1 plots, in Immokalee and Citra, respectively. In Immokalee, the CSF provided the most significant weed control, but ASD treatments also suppressed weeds enough to prevent an impact on yield. In Citra, all treatments, including the CSF, provided poor weed control relative to the Immokalee site. In both locations, the application of ASD provided a level of root-knot nematode (*Meloidogyne* sp.) control equivalent to, or more effective than the CSF. In Immokalee, ASD2 and ASD1 plots provided 26.7% and 19.7% higher total marketable yield as compared with CSF plots, respectively. However, in Citra, total marketable yield was unaffected by soil treatments. Tomato fruit quality parameters were not influenced by soil treatments, except for fruit firmness in Immokalee, which was significantly higher in fruits from ASD treatments than in those from CSF soil. Fruit mineral content was similar or higher in ASD plots as compared with CSF. In fresh-market tomato, ASD applied using a mixture of CPL and molasses may be a sustainable alternative to CSF for maintaining or even improving marketable yield and fruit quality.

Soilborne fungal pathogens, nematodes, and weeds represent some of the most important biotic factors limiting vegetable crop production and profitability in the world. After the phaseout of methyl bromide, although other chemical soil fumigants (CSF) are available, there is still a pressing need for effective, viable, and more sustainable options (Shennan et al., 2014). Among the nonchemical alternatives, ASD, also known as “biological soil disinfestation” (Blok et al., 2000) or “reductive soil disinfestation” (Shimura et al., 1999), is considered as one of the most promising methods. Anaerobic soil disinfestation has proved to be effective against several soilborne fungal and bacterial plant diseases, and plant-parasitic nematodes and weeds, across a wide range of crops and environments (Butler et al., 2012a, 2012b; Lamers et al., 2010; Momma, 2008; Roskopf et al., 2015; Shennan et al., 2014). Developed independently in Japan (Shimura et al., 1999) and in the Netherlands (Blok et al., 2000), for both open field and protected crops, the technique is gaining interest in the United States, China, and other countries (Kim et al., 2007; Meng et al., 2015; Shennan et al., 2014). Suitable also for raised-bed crops, ASD does not require the use of chemicals and may be applied even in organic production systems. The current approach to ASD treatment in Florida recommends application 3 weeks before crop transplanting and consists of creating temporary anaerobic (reducing) conditions by 1) amending the soil with a readily decomposable C source to initiate rapid soil microbial growth and respiration, 2) covering the bed with oxygen impermeable polyethylene mulch to minimize gas exchange, and 3) irrigating the soil to saturate the pore space, which, besides creating anaerobic conditions, enhances the diffusion of by-products through the soil solution within the volume of soil that will host the crop root system (Butler et al., 2014; Shennan et al., 2014).

The growth of aerobic microorganisms stimulated by the organic amendment causes a rapid decline in oxygen content in the soil with consequent decrease of the redox potential (Eh) and the development of anaerobic conditions that promote the growth of facultative and obligate anaerobic microorganisms over the aerobic microbial community (Mowlick et al., 2012, 2013a, 2013b; van Agtmaal et al., 2015).

Under reducing conditions, the organic matter is subject to fermentation with consequent production of short-chain fatty acids (acetic, butyric, and propionic acids), aldehydes, alcohols, and volatile organic compounds (VOC) that are toxic and/or suppressive for several soilborne pathogens, plant-parasitic nematodes, and weeds (Bonanomi et al., 2007; Momma et al., 2006; Momma, 2008; Oka, 2010; van Agtmaal et al., 2015). Momma et al. (2011) reported that in presence of anaerobic conditions the generation of ions such as Fe²⁺ and Mn²⁺ may contribute to the suppression of soilborne pathogens like *Fusarium oxysporum*.

Although the mechanism of pest suppression by ASD is not fully understood and

requires further research, an aspect that needs particular attention is the ASD application technique. It is critical for the adoption of ASD at the commercial level, to define and validate a feasible field-scale ASD application procedure. The main factors affecting the level of anaerobiosis and low pH achievable, as well as the microbial type and population growth, and the maintenance of reducing conditions overtime are the soil type, the initial water volume applied, and the type and rate of organic matter applied (Butler et al., 2012b, 2014). A good C source should be locally and abundantly available, low cost, homogenous, easy to apply, and effective in supporting microbial growth. Depending on local availability and costs, the C source may be constituted by cover crop residues, ethanol, molasses, rice or wheat bran, or a combination of organic materials (Rosskopf et al., 2015; Shennan et al., 2014).

Although temporary, the reduction of Eh and the lowering of pH caused by the ASD treatment (Momma, 2008), may have a substantial impact on the pests and the entire soil-microorganism-plant system (Husson, 2013; Strauss and Kluepfel, 2015; van Agtmaal et al., 2015). Combined with solarization and using molasses as a C source, the ASD treatment provided equivalent or greater marketable yields than the methyl bromide control in a bell pepper (*Capsicum annuum* L.)—eggplant (*Solanum melongena* L.) double crop system (Butler et al., 2014). However, while great attention has been given to the efficacy of control of the ASD against specific pests and pathogens (Blok et al., 2000, 2012a, 2012b; Lamers et al., 2010; Momma, 2008; Shennan et al., 2014), little is known about the effects of the ASD per se on tomato (*S. lycopersicum*) crop yield and fruit quality.

Therefore, a field study was carried out in two different locations in Florida, which leads the United States in the production of fresh-market tomato, accounting for 34% of the U.S. fresh-market tomato harvested area and 39% of the national crop value in 2014 (USDA-NASS, 2015), the majority of which is produced using soil fumigation as the basis for pest management. The study was conducted in an open-field, fresh-market tomato production system to evaluate ASD in comparison with the reference CSF treatments for weeds and nematodes control as well as for influence on fruit yield and quality.

Materials and Methods

Experimental sites and treatments. During the spring season of 2015, two experiments were conducted in open-field fresh-market tomato in southwestern (Immokalee) and northern Florida (Citra). The conventional

CSF was compared with two ASD treatments, which consisted of amending the soil with 22 Mg·ha⁻¹ of CPL and two rates of molasses [13.9 (ASD1) and 27.7 m³·ha⁻¹ (ASD2)] as a C source. The first experiment was established on 2 Feb. 2015 at the University of Florida (UF)/Institute of Food and Agriculture Science (IFAS)/South West Florida Research and Education Center (SWFREC) in Immokalee, FL. The second experiment was established in North Florida on 25 Mar. 2015, at the UF Plant Science Research and Education Unit in Citra, FL.

In Immokalee, the soil was a Spodosol classified as Immokalee fine sand (sandy, siliceous, hyperthermic Arenic Haplaquods), and the experimental field was previously characterized as having moderate weed and root-knot nematode pressure. In Immokalee, weed species homogeneously infesting the experimental field included yellow nutsedge (*Cyperus esculentus* L.), the monocotyledonous goosegrass [*Eleusine indica* (L.) Gaertn.], southern crabgrass [*Digitaria ciliaris* (Retz.) Koel.], large crabgrass [*Digitaria sanguinalis* (L.) Scop.], and smooth crabgrass [*Digitaria ischaemum* (Schreb.) Muhl.], and the dicotyledonous pigweed (*Amaranthus retroflexus* L.), false daisy (*Eclipta prostrata* L.), and old world diamond (*Hedyotis corymbosa* L.), which represent some of the most common weeds infesting tomato crops in the area. In Citra, the soil was a Gainesville loamy sand (hyperthermic, coated typic quartzipsamments) and the experimental field had high weed pressure, consisting predominantly of yellow nutsedge. At this site, root-knot nematode pressure was also high based on previous observations.

At each location, treatments (CSF, ASD1, and ASD2) were arranged in a randomized complete block design with four replications. A nontreated control was not included because the ASD method has been proven to have efficacy against multiple soilborne pest species when compared with untreated soil (McCarty et al., 2014), while the comparison with the current commercial fumigants available in each area is critical for the adoption of ASD at commercial level. In Immokalee, each of the four blocks, was constituted by one raised bed, 0.90 m wide, 0.20 m high, and 60 m long, and treatments were applied to 15-m-long sections of the bed, with 3-m space between plots. In Citra, each of the four blocks consisted of three beds 0.90 m wide and 15-m long. In both locations, before treatment application, soil was rototilled and a starter fertilizer mix including nitrogen (N), phosphorous (P), and potassium (K) was applied at the rate of 34, 49, and 37 kg·ha⁻¹ at the Immokalee site, and 56, 22, and 42 kg·ha⁻¹ at the Citra site, respectively. In both locations, the starter fertilizer mix was broadcast applied to the soil surface on a band of 60 cm wide. Then, rounded false beds were formed hilling the soil from a depth of 10 cm, and ASD beds were amended with CPL at the rate of 22 Mg·ha⁻¹, and with a 1:1 (v:v) water dilution of sugarcane molasses (agricultural carbon source, Terra Feed, LLC, Plant

City, FL). The CPL contained 23.0% of C, 2.6% of N, 1.4% of P, and 2.5% of K. Molasses had a density of 1420 kg·m⁻³, 22% of water content, a pH of 4.9–5.2, 1.23% of N and 34.25% of C. The molasses–water mix was applied to ASD1 and ASD2 plots at the rate of 27.7 and 55.4 m³·ha⁻¹.

After CPL and molasses application, the soil was tilled to a depth of 15 cm with a rotary cultivator, beds were formed and covered with a 0.03-mm black/white VaporSafe® TIF (Raven Industries Inc., Sioux Falls, SD) polyethylene mulch containing an ethylene vinyl alcohol barrier layer. Simultaneously, two drip irrigation lines [20 cm emitter spacing, 0.98 L·h⁻¹ emitter rate (Jain Irrigation Inc., Haines City, FL)] were installed under the mulch in each bed, ≈2.54 cm below the soil surface and 20 cm apart from the center of the bed.

ASD plots were then irrigated for ≈4 h at the rate of 5 cm of water (based on raised-bed area only) to saturate air-filled pore space in the top 10 cm of the bed and enhance the development of anaerobic conditions (Butler et al., 2012a).

On the same day, the control plots were fumigated by shank injection with Pic-Clor 60 (Soil Chemical Corporation, Hollister, CA) containing a mixture of 1,3-dichloropropene (39.0%) and chloropicrin (59.6%) at the rate of 224 kg·ha⁻¹ in Immokalee, and with Paladin™ (Arkema Inc., King of Prussia, PA) composed of DMDS (79%) and chloropicrin (21%) at the rate of 496 L·ha⁻¹ in Citra. In Immokalee, CSF was performed to a depth of 23 cm with three shanks; external shanks were 56 cm apart, and a third shank was placed in the middle off-centered, 23 and 33 cm apart from the external shanks. In Citra, CSF was performed to a depth of 25 cm with two shanks placed 30 cm apart. In both locations, fumigated plots were mulched right after CSF.

Soil pH and redox potential. Before initial irrigation, two oxidation–reduction potential sensors (Pt combination electrodes, Ag/AgCl reference; Sorex, Garden Grove, CA) were installed to a depth of 15 cm in each plot, to measure the redox potential (Eh), and thus, to evaluate the level of anaerobiosis achieved in the soil during the first 3 weeks after treatment application (WATA) application. Electrodes were continuously monitored using an automatic data logging system (CR-1000 with AM 16/32 multiplexers; Campbell Scientific, Logan, UT) and the collected data were used to calculate the cumulative number of hours under anaerobic conditions. Raw soil redox potential values were corrected to relate to the redox potential of a standard hydrogen electrode (Fiedler et al., 2007). Hourly average soil redox potential values below the calculated critical redox potential (CEh) were considered to be indicative of anaerobic conditions. Critical redox potential was considered as an indicator of reduced (anaerobic) soil conditions by the USDA-NRCS (Rabenhorst and Castenson, 2005; USDA-NRCS, 2010), and was calculated using the formula:

$$CEh = [595(\text{mV}) - 60(\text{mV})] \times \text{soil pH}$$

Soil pH was measured three WATA to calculate CEh. For values below CEh, the

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absolute value of the difference between a given value and CEh were summed over the 3-week treatment period to provide a measure of cumulative soil anaerobic conditions. Using a soil probe (1.75-cm internal diameter), six soil cores were taken randomly from each plot at 20-cm depth and combined in a bulk sample for each plot. Soil pH was measured directly in each soil sample (without an extraction) using a FieldScout SoilStik pH meter (Spectrum Technologies, Inc., Aurora, IL).

Crop transplanting and growing conditions. Tomato plants of 'Skyway 687' (Enza Zaden, Salinas, CA) in Immokalee and 'Tribute' (Sakata, Morgan Hill, CA) in Citra, both large, round fresh-market commercial tomato varieties, were transplanted at the third true leaf stage, three WATA on 24 Feb. and 17 Apr. 2015 in Immokalee and Citra, respectively. Both varieties were selected for their intermediate resistance to *Tomato spotted wilt virus* and *Tomato yellow leaf curl virus*. 'Skyway 687' is also resistant to *Tomato apex necrotic virus*, fusarium wilt, verticillium wilt, and has intermediate resistance to root-knot nematodes, whereas 'Tribute' is resistant to fusarium wilt, verticillium wilt, and alternaria stem canker, with intermediate resistance to gray leaf spot. In both experiments, tomato plants were planted in single rows, at a distance of 0.45 m within the row and 1.8 m between rows, establishing a density of 12,000 plants/ha. Each of the 12 plots contained 34 plants.

In both locations, plants were trellised using the stake and weave method. In Immokalee, the crop was managed using a hybrid seepage drip irrigation system, whereas in Citra, the crop was watered by drip irrigation. In Immokalee, irrigation volumes were defined daily based on the Florida Automated Weather Network (FAWN) recommendations for tomato. While in Citra, irrigation volumes were timer controlled with a run time of 30 min twice a day. Soil moisture in the root zone was monitored by tensiometers installed in each replication. In Immokalee, the water table was regulated on a daily basis by monitoring wells installed at both ends of the external beds. The water table was maintained between 40 and 80 cm below the bed surface. The monitoring wells were constructed of 1.2-m-long 10-cm-diameter poly vinyl chloride (PVC) pipes screened 20 cm from the bottom (Smajstrla and Muñoz-Carpena, 2011). A float was attached to one end of a PVC pipe to serve as the water level indicator. Permanent marks were made every 25 mm to indicate the water table depth below the polyethylene mulch bed.

Fertigation was started 3 weeks after planting following the UF/IFAS fertilizer recommendations (FDACS, 2005). At the Immokalee site, N and K were applied twice a week by fertigation using potassium nitrate (13–0–44) and ammonium nitrate (34–0–0). In Citra, fertilizer was applied by fertigation once per week using a 6–0–8 plus micro blend by Mayo Fertilizer Inc. (Mayo, FL). Total in-season fertilizer rates applied by fertigation were 180 and 268 kg·ha⁻¹ of N

and 263 and 265 kg·ha⁻¹ of K in Immokalee and Citra, respectively. Total N, P, and K fertilizer rates were 214, 49, and 300 kg·ha⁻¹ in Immokalee, and 323, 22, and 307 kg·ha⁻¹ in Citra, respectively.

Weather data were obtained by the FAWN (<http://fawn.ifas.ufl.edu/>) stations located in Immokalee and Citra. Pests and diseases were managed following the UF/IFAS recommendations based on weekly scouting (Freeman et al., 2014). In Citra, the crop was uniformly attacked by early blight (*Alternaria solani*); despite the regular application of fungicides, the disease severely affected the crop, thereby affecting fruit yield.

Weed evaluation, soil sampling, and nematode analysis. The number of emerging weeds and the percentage of weed coverage of each plot were evaluated from four subsamples (0.25 m²) at 9, 22, 37, 50, 69, and 80 DAP in Immokalee, and at 20, 81, and 95 DAP in Citra. For nematode analysis, soil samples were collected, as previously described for soil pH analysis, before treatment initiation, at transplanting, and at the end of the crop season in both locations (80 DAP in Immokalee, and 91 DAP in Citra). A subsample of 100-cm³ soil was used to extract nematode second stage juveniles (J2) using the Baermann funnel technique. Nematodes were counted and identified as root-knot *Meloidogyne* sp. or nonparasitic using inverted microscopes. At the end of the season, three randomly selected plants were removed from each plot and the roots were rated for galling and root disease. Galling was assessed on a scale of 0 to 10, with 10 representing severe (100%) galling (Bridge and Page, 1980). A subjective scale of 0 to 5 was used to assess root disease, with 0 representing healthy roots with no apparent signs of damage and 5 representing completely diseased and degraded roots. Root-knot nematode J2 were extracted from plant root tissue by placing a subsample of root tissue into funnels for ≈60 h, after which root-knot nematode J2 were identified and counted microscopically, as described previously.

Fruit yield and grade distribution. Yield was measured from one 4.5-m long (10 plants) representative section in each experimental unit. Fruits ranging from marketable mature green to ripe were harvested three times on 29 Apr., 13 and 26 May 2015 [64, 78, and 91 d after planting (DAP)], and on 26 June, 6, and 17 July 2015 (70, 80, and 91 DAP) in Immokalee and Citra, respectively. Fruits were graded into size categories extra-large (greater than 7.00 cm), large (6.35 to 7.06 cm), medium (5.72 to 6.43 cm), and unmarketable fruit according to USDA grade standards (USDA, 1991) and weighed.

Postharvest fruit quality. In both locations, at the first harvest, a subsample of 20 mature-green tomato fruit per plot was collected. In Immokalee, samples were placed in labeled paper bags, and transported to the Gargiulo, Inc. packing house (Immokalee, FL), where fruit were subject to ethylene treatment at 20 °C and 85% to 90% relative humidity until Stage 2 of ripeness (breaker)

(Sargent et al., 2005; USDA Agriculture Marketing Service, 1997). After tomatoes achieved Stage 2, they were transported to the Vegetable Laboratory (UF/IFAS/SWFREC) and ripened at room temperature (23 to 24 °C) until the table-ripe stage or Stage 5 to 6 for quality evaluations. In Citra, fruit samples were allowed to ripen at 25 °C until table-ripe stage. Fruit firmness was measured on four fruits per plot as fruit deformation using an 11-mm probe and 1-kg force applied to the fruit equator area for 5 s using a portable digital firmness tester (Model C125EB; Mitutoyo, Corp., Aurora, IL) for the Immokalee trial and the TA.HD Plus Texture Analyzer (Texture Technologies Corp and by Stable Micro System, Ltd. Hamilton, MA) for the Citra trial. Exterior fruit color was measured on the same four fruit using a 1 to 6 scale where 1 = green and 6 = red (USDA, 1997). Total soluble solids (TSS) and pH analysis were performed using one-fourth of each of the four fruit. For each tomato fruit sample, ≈200 g (fresh weight) of fruit was dried in a forced-air oven at 65 °C for 72 h and then weighed to calculate the fruit dry matter (dry water) content.

Mineral analysis. Dried fruit samples were finely ground through a mill to pass through a 20-mesh screen, then 0.5 g of the dried plant tissues were analyzed for the following elements: N, P, K, Ca, Mg, Fe, B, Cu, Zn, Mn, Na, Zn, Mo, and Ni. Total N content was determined by dry combustion (NC Soil Flash EA1112; CE Elantech Inc., Lakewood, NJ). Dry fruit samples were digested using a closed-vessel microwave-assisted digestion (MARS Express; CEM Corp., Matthews, NC) according to U.S. EPA method 3052 (USEPA, 1997). Digested products were then analyzed by inductively coupled plasma atomic emissions spectrometry (ICP-AES; iCAP 6500, Thermo Scientific, Waltham, MA) to determine the concentrations of P, K, Ca, Mg, Fe, B, Cu, Zn, Mn, Na, Zn, Mo, and Ni.

Statistical analysis. Data from the two locations were analyzed separately. Collected data on cumulative Eh, weed number and coverage, nematode population, fruit yield, and quality were subject to analysis of variance (ANOVA) using the GLM procedure in SAS Version 9.2 software (SAS Institute, Cary, NC). All means were separated using Duncan's multiple range test at $P = 0.05$. Weed cover percentage data were transformed by arcsine transformation before ANOVA to obtain a normal distribution.

Results and Discussion

Weather conditions. In Immokalee, mean daily air temperature was on average 16.1 °C during the 3-week treatment and 23.9 °C during the crop season (from transplanting until final harvest). During the first three WATA, daily minimum and maximum air temperatures ranged from -0.9 to 15.5 °C and 15.5 to 29.6 °C, respectively. During the cropping season, daily minimum and maximum air temperature ranged from 8.2 to 22.2 °C and 18.5 to 34.6 °C, respectively

(Fig. 1). A single freezing ($-0.89\text{ }^{\circ}\text{C}$) event was recorded 4 d before crop transplanting, while after crop transplanting temperatures never dropped below $8\text{ }^{\circ}\text{C}$. After crop transplanting, daily solar irradiance was on average $215.6\text{ W}\cdot\text{m}^{-2}$ and cumulative precipitation was 287 mm. Based on the definition of leaching rain (76 mm of rain in 3 d or 102 mm in 7 d) provided by Simonne and Hochmuth (2011), one leaching rainfall event with 82 mm of rainfall was recorded in Immokalee on 12 May (77 DAP) just before the second harvest.

In Citra, mean daily air temperature was $20.8\text{ }^{\circ}\text{C}$ during the three WATA and $25.1\text{ }^{\circ}\text{C}$ during the entire cropping season after transplanting. In the 3-week treatment, daily minimum and maximum air temperature ranged from 3.7 to $20.3\text{ }^{\circ}\text{C}$ and 18.9 to $30.8\text{ }^{\circ}\text{C}$, respectively. After crop transplanting, during the cropping season, daily minimum and maximum air temperatures ranged from 8.7 to $24.7\text{ }^{\circ}\text{C}$ and 24.5 to $36.9\text{ }^{\circ}\text{C}$, respectively (Fig. 1). No freezing events were recorded during the season. After transplanting the crop, daily solar irradiance was, on average $230.2\text{ W}\cdot\text{m}^{-2}$ and cumulative precipitation was 318.3 mm . A leaching rainfall event with 83 mm of rainfall also occurred in Citra and was recorded on 8 July (82 DAP). Cumulative precipitation recorded in both locations was within the range of mean cumulative rainfall typically recorded in Florida during the spring season (Fraisse et al., 2010).

Soil redox potential. In both locations, plots subjected to ASD showed significantly higher cumulative anaerobic conditions as compared with the CSF plots that remained aerobic (Fig. 2). The level of cumulative anaerobiosis was significantly influenced by the molasses rate. The level in ASD2 plots was 167% and 116% higher than in ASD1 plots in Immokalee and Citra, respectively (Fig. 2). These results suggest that the level of anaerobic conditions achievable in the soil is significantly influenced by the amount of C source used to amend the soil. Moreover, these results confirm the effectiveness of the ASD technique in establishing anaerobic conditions in the soil, which is consistent with the findings by Butler et al. (2012a) in which an increase in anaerobiosis was observed in soil amended with molasses and CPL. In addition, the anaerobic levels achieved in the experiment conducted by Butler et al. (2012a) confirmed that even with soil temperatures averaging $25\text{ }^{\circ}\text{C}$ during the ASD application, opaque TIF plastic was effective in reaching threshold cumulative Eh levels, similar to levels reported with solarization film.

Soil treatment effects on weeds. After the phaseout of methyl bromide, weed control became one of the major issues in the vegetable industry, as none of the soil disinfection alternatives currently available has shown consistent results and can assure a complete weed control in all the application conditions. The effectiveness of several CSFs, currently available is in fact, influenced by the specific application conditions (soil type, soil moisture, infestation level, bed geometry,

application equipment). Therefore, also for the ASD technique, it was critical to evaluate the efficacy of control against weeds, at field level, under specific application conditions, and in comparison with different CSF reference standards.

In Immokalee, soil treatments exerted a significant effect on both weed number and coverage percentage (Table 1). Since the first assessment at 9 DAP and during the entire crop cycle, CSF assured a near complete control of all weeds. Plots subject to ASD1 showed the highest weed number, ranging on average from 9.25 to 8.25 shoots/ m^2 at 9 and 80 DAP, respectively, as well as the highest weed coverage percentage, ranging from an average of 0.03% at 9 DAP to 16.25% at 80 DAP. However, no differences were observed between plots treated with ASD1 and ASD2 throughout the season, both in terms of weed number and percent coverage. Weeds developed mostly through planting holes and only a few yellow nutsedge shoots were able to penetrate the TIF polyethylene mulch. Moreover, it was observed that the application of ASD especially at higher molasses rate (ASD2) substantially reduced the emergence of weeds in the alleys among beds, which remained clean until first harvest. This could be explained by the partial movement of the molasses outside the bed. At the end of the season, considering the percent of weed coverage, CSF displayed an efficacy approaching 100%, while ASD treatments showed an average efficacy of 85%.

In Citra, all soil treatments including the CSF resulted in a very low level of weed control although there were no significant differences among treatments (Table 1). The number of emerging weeds ranged from an average of 13.2 shoots/ m^2 at 20 DAP to 37.7 and 26.3 shoots/ m^2 at 81 and 91 DAP, respectively (Table 1). Weed percent coverage ranged from 14.7% at 20 DAP to 59.8% at 91 DAP, and was not influenced by soil treatments, except at 81 DAP, when ASD2 plots showed a percent weed coverage lower than ASD1, 31.3% vs. 54.4% ($P = 0.05$). In plots with CSF, the percent weed coverage was equivalent to that of ASD plots regardless the applied molasses rate.

In presence of high nutsedge pressure, such as that observed in Citra, neither ASD nor DMDS provided acceptable weed control. After the loss of methyl bromide, increased research on alternative fumigants has established the need for an herbicide partner with the majority of alternatives, particularly where nutsedge is the principal weed problem (Noling et al., 2006). However, where grass weeds dominate or under relatively low weed pressure, as observed in Immokalee, the application of ASD can provide adequate weed control, as reflected in increased crop yield (see below). Considering the limited weed control efficacy provided by the ASD in Citra, it may be interesting to investigate in future studies the possibility to combine the ASD technique with the application of herbicides.

Soil treatment effects on plant-parasitic and nonparasitic nematodes. To evaluate the

impact of soil disinfection treatments on soil nematode populations, both plant-parasitic and nonparasitic nematodes were analyzed immediately before treatment application, at three WATA and at the end of the crop season. Before treatment application, the plant-parasitic nematode population was low in Immokalee, where the root-knot nematode population was on an average of 2.84 J2 per 100 cm^3 of soil, while it was relatively high in Citra with an average root-knot nematode population of 10.40 J2 per cm^3 of soil. The number of nonparasitic nematodes was on average 238 per cm^3 of soil in Immokalee and 491 per cm^3 of soil in Citra (Table 2). All soil treatments reduced the root-knot nematode population to zero at three WATA in both locations (Table 2), demonstrating the effectiveness of ASD in controlling root-knot nematodes. These results were consistent with the findings of Butler et al. (2012a) who observed a decrease of root-knot nematodes in plots amended with molasses and/or CPL in a pepper-eggplant double crop system. The control of root-knot nematodes in ASD plots may be explained by the nematotoxic effect of organic acids and ammonium released during the decomposition of the organic matter (Katase et al., 2009; Thoden et al., 2011). At the same time, in Immokalee, the nonparasitic nematodes population increased up to 2840 per cm^3 of soil in ASD2 plots, and was significantly higher ($P = 0.004$) in soil treated with ASD at both molasses rates than in CSF soil, where even the nonparasitic nematode population was reduced to zero. Whereas in Citra, all tested treatments reduced the population of nonparasitic nematodes, and although ASD plots showed higher levels of nonparasitic nematodes, no significant difference ($P = 0.07$) was observed between ASD and CSF plots (Table 2). The increase of nonparasitic nematodes observed in Immokalee only in ASD plots, may be due to the soil amendment with CPL, and was consistent with the findings of several authors, who have observed that the number of free-living bacterial- and fungal-feeding nematodes markedly increases after the addition of any form of organic soil amendment (Thoden et al., 2011).

At the end of the season, in Immokalee, the root-knot nematode population remained low, ranging from an average of 0 J2 per cm^3 of soil in ASD2 plots to 17 J2 per cm^3 of soil in plots subject to ASD1, but was not significantly ($P = 0.36$) different among the treatments. Also in Citra, the end-of-season root-knot nematode population was not significantly ($P = 0.54$) different among the treatments; however, in this case, the parasitic nematode population increased in all treatments and was on average of 44.4 J2 per cm^3 of soil (Table 2). The root-knot nematode population increase observed in Citra at the end of the season could be at least in part due to the low weed control obtained in this location, as weeds may host parasitic nematodes (Rich et al., 2009). Such results suggest the importance of controlling both weed and nematodes at the same time. In both locations, the end-of-season nonparasitic nematode population

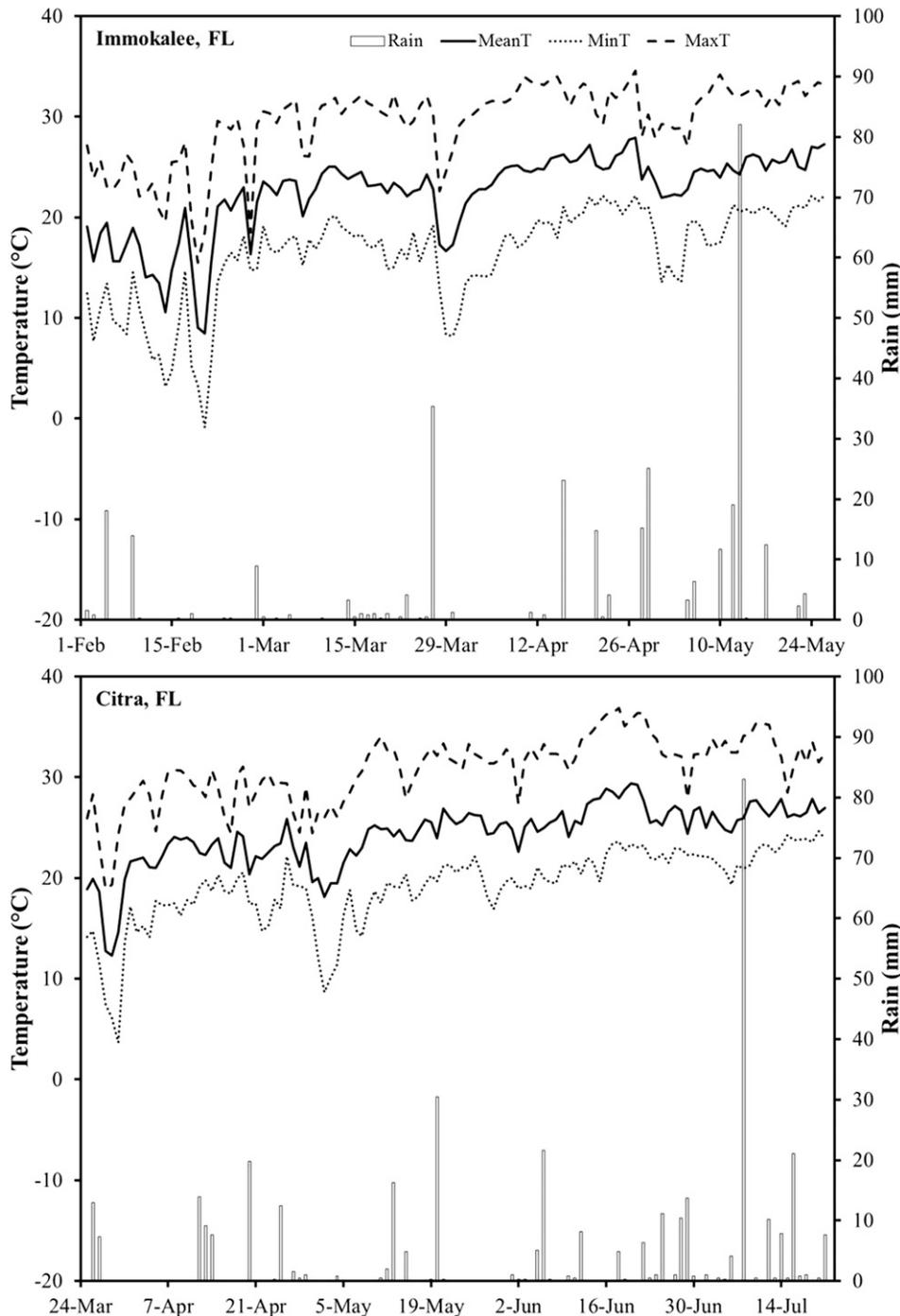


Fig. 1. Mean, minimum, maximum temperatures and rainfall recorded during the crop cycle in Immokalee (University of Florida/Institute of Food and Agricultural Science/Southwest Florida Research and Education Center) and Citra (University of Florida/Plant Science Research and Education Unit) in the spring of 2015. Weather data obtained from the Florida Automated Weather Network station located at the SWFREC in Immokalee and at the Plant Science Research and Education Unit in Citra, FL.

was not significantly ($P = 0.50$ and 0.12) different among the treatments.

At the end of the crop season, an assessment of the nematode population and root condition ratings were also performed on the tomato roots (Table 3). While the number of root-knot nematode J2 per gram of roots was not significantly different among the treatments ($P = 0.12$ in Immokalee and 0.31 in Citra), the root gall index was significantly lower ($P = 0.01$) from roots subjected to ASD1 than from those

subjected to ASD2 or CSF in Immokalee. In Citra, the root gall index was significantly lower ($P = 0.04$) on roots from ASD1 plots than in those subjected to CSF. As expected, based on initial numbers in Immokalee, the number of root-knot nematodes per gram of root was low ranging from a minimum average of 0.56 J2/g of root in ASD1 to 2.05 J2/g of roots in CSF plots. In Citra, the number of root-knot nematodes per gram of root was higher and ranged from a minimum average of 5.45 J2/g of

roots in ASD1 to a maximum average of 13.24 J2/g of roots in ASD2 plots. In both locations, the number of nonparasitic nematodes per gram of roots was not significantly different among the treatments. It follows that ASD can assure a level of control of the root-knot nematode population equivalent or higher than that provided by the CSF. Moreover, regardless of the molasses rate, the ASD has a lower impact on the nonparasitic nematode population in soil compared with the CSF, thus maintaining

a higher level of soil biodiversity, even though it did not last for the entire cropping season.

Soil treatment effects on fruit yield and grade distribution. In Immokalee, the soil disinfestation treatments tested showed no influence on total marketable yield at first harvest and first and second harvest combined (FSHC). However, total season marketable yield was significantly ($P = 0.03$) higher in ASD than CSF plots (Table 4). With

a total season marketable yield of 62.1 Mg·ha⁻¹, plots subject to ASD2 provided the highest tomato fruit yield, which did not differ from the fruit yield obtained at lower molasses rate (ASD1). Compared with the CSF, ASD2 and ASD1 plots provided 26.7% and 19.7% higher total season marketable yield, respectively. Total marketable yields obtained in Immokalee were within the range of those reported by Ozores-Hampton et al. (2012, 2015) for the cultivar Florida 47 R

grown in southwestern Florida during the spring (51.8–109.5 Mg·ha⁻¹). Extra-large fruits declined overtime from 82.5% at first harvest to 70.2% of the total season marketable yield; however, soil disinfestation treatments had no significant effect on the fruit size distribution either at first harvest, FSHC, or total season harvest. Soil treatments did not influence the amount of total unmarketable fruit, which averaged 7.6% of the total season fruit yield.

Given the relatively low weed and nematode pressure observed in the Immokalee site, the higher total season marketable yield measured in ASD plots as compared with CSF may be explained by the higher water and nutrient-holding capacity conferred to the soil by the CPL (Butler et al., 2014; Ozores-Hampton et al., 2011), rather than by improved control of root-knot nematodes and weeds.

In Citra, total marketable yield was on average 11.8 Mg·ha⁻¹, resulting in lower than normal yield range (55–78 Mg·ha⁻¹) for the same area and season (Zotarelli et al., 2009), and was not significantly ($P = 0.19$) affected by soil disinfestation treatment (Table 4). Total marketable yield was not significantly influenced by soil treatments also at first harvest ($P = 0.47$) and at FSHC ($P = 0.30$). At first harvest, extra-large, large, and medium sized fruits accounted on average for 48.9%, 38.1%, and 12.9% of the total marketable yield, respectively. At the end of the season, 36.8% of the marketable fruits were large, and extra-large and medium sized fruit accounted for 31.6% and 31.7% of the total season marketable yield. However, also in Citra, fruit size distribution was not significantly affected by treatments either at first harvest, FSHC, or total season harvest. The amount of unmarketable fruits was on average 28.8% of total season fruit yield and was not influenced by soil disinfestation treatments (Table 4).

The low total marketable yield obtained in Citra, regardless of soil treatment, may be attributable to the combined effect of the high weed and root-knot nematode pressure, as well as to the later than average planting date and to the occurrence of early blight that had a detrimental impact in all of the treatments in all of the replications. As Stall and Morales-Payan (2003) found that season-long interference of 25 yellow nutsedge plants per meter square resulted in a 10% reduction in marketable tomato yield, probably over 10% of the yield reduction observed in Citra could be due only to the nutsedge competition.

In fresh-market tomato, the use of ASD can provide equal or higher marketable yields as compared with a standard CSF, which is consistent with the findings of Butler et al. (2014) who observed good yield performances combining ASD and solarization for production of bell pepper and eggplant.

Soil treatment effects on fresh-market tomato fruit quality and mineral content. In Immokalee, tomato fruit color, TSS, pH, and DM content were not influenced by the soil disinfestation treatments, while a significant

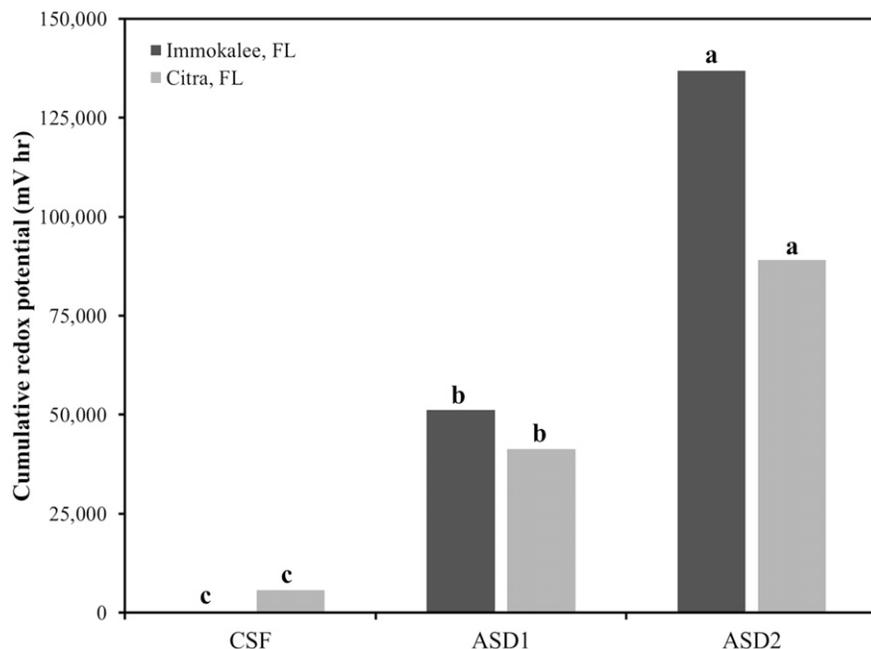


Fig. 2. Soil treatment effect on mean cumulative soil redox recorded in the 3 weeks after treatment application in Immokalee (University of Florida/Institute of Food and Agricultural Science/Southwest Florida Research and Education Center) and Citra (University of Florida/Plant Science Research and Education Unit) in the spring of 2015. Where CSF (chemical soil fumigation, with Pic-Clor 60 at the rate of 224 kg·ha⁻¹ in Immokalee, and Paladin™ at the rate of 496 L·ha⁻¹ in Citra), ASD1 [anaerobic soil disinfestation with 13.9 m³·ha⁻¹ of molasses, and 22 Mg·ha⁻¹ of composted poultry litter (CPL)], ASD2 (anaerobic soil disinfestation with 27.7 m³·ha⁻¹ of molasses, and 22 Mg·ha⁻¹ of CPL). Different letters, within each location, indicate significant differences at $P = 0.05$ by Duncan's multiple range test.

Table 1. Soil treatment effects on weed count and coverage of fresh-market tomato beds mulched with totally impermeable film grown in Immokalee (University of Florida/Institute of Food and Agricultural Science/Southwest Florida Research and Education Center) and in Citra (University of Florida/Plant Science Research and Education Unit) in the spring of 2015.^z

DAP ^y	Weed count (number/m ²)			P value	Weed coverage (%)			P value
	CSF ^x	ASD1	ASD2		CSF	ASD1	ASD2	
Immokalee, FL								
9	0.50 c	9.25 a	5.50 b	0.002	0.0	0.0	0.0	0.42
22	1.00 b	8.20 a	5.80 a	0.01	0.0	0.3	0.1	0.24
37	0.00 b	8.60 a	6.00 a	0.01	0.1 b	4.5 a	3.0 a	0.02
50	0.00 b	7.75 a	4.50 ab	0.02	0.1 b	6.3 a	6.8 a	0.04
69	0.00 b	7.50 a	5.50 a	0.001	0.1 b	12.5 a	10.5 a	0.02
80	0.00 b	8.25 a	6.50 a	0.001	0.1 b	16.3 a	13.0 a	0.04
Citra, FL								
20	11.75	14.25	13.50	0.88	13.1	19.3	11.9	0.46
81	38.50	45.00	29.50	0.09	43.8 ab	54.4 a	31.3 b	0.05
91	22.00	27.50	29.50	0.34	63.1	59.4	56.9	0.91

^zWeed evaluations were made from four subsamples (0.25 m²) per treatment. Reported values are averages of four replications. Means followed by different letters within each assessment date (row) and experiment are significantly different at $P = 0.05$ by Duncan's multiple range test.

^yDAP = Days after planting.

^xCSF (chemical soil fumigation, with Pic-Clor 60 at the rate of 224 kg·ha⁻¹ in Immokalee, and Paladin™ at the rate of 496 L·ha⁻¹ in Citra), ASD1 [anaerobic soil disinfestation with 13.9 m³·ha⁻¹ of molasses, and 22 Mg·ha⁻¹ of composted poultry litter (CPL)], ASD2 (anaerobic soil disinfestation with 27.7 m³·ha⁻¹ of molasses, and 22 Mg·ha⁻¹ of CPL).

Table 2. Soil root-knot and nonparasitic nematode juveniles (J2) counted before and after treatment application in Immokalee (University of Florida/Institute of Food and Agricultural Science/Southwest Florida Research and Education Center) and Citra (University of Florida/Plant Science Research and Education Unit) in the spring of 2015.^z

Assessment timing	Treatments ^y	Root-knot nematodes (J2/cm ² soil)	Nonparasitic nematodes (number/cm ² soil)	
Immokalee, FL	Pretreatment	2.84	238.14	
	Posttreatment (21 DAT) ^x	<i>P</i> value	—	—
		CSF	0.00	0.00 b
		ASD1	0.00	2,098.00 a
		ASD2	0.00	2,840.80 a
	Harvest	<i>P</i> value	—	0.004
		CSF	2.84	209.75
		ASD1	17.01	572.75
		ASD2	0.00	303.25
	Citra, FL	Pretreatment	10.40	491.40
Posttreatment (21 DAT)		<i>P</i> value	—	—
		CSF	0.00	27.00
		ASD1	0.00	138.50
		ASD2	0.00	129.25
Harvest		<i>P</i> value	—	0.07
		CSF	28.35	164.43
		ASD1	62.37	172.94
		ASD2	42.53	725.76
		<i>P</i> value	0.54	0.12

^zReported values are averages of four replications. Means followed by different letters within a column for each experiment and assessment timing are significantly different at $P = 0.05$ by Duncan's multiple range test.

^yCSF (chemical soil fumigation, with Pic-Clor 60 at the rate of 224 kg·ha⁻¹ in Immokalee, and Paladin™ at the rate of 496 L·ha⁻¹ in Citra), ASD1 [anaerobic soil disinfestation with 13.9 m³·ha⁻¹ of molasses, and 22 Mg·ha⁻¹ of composted poultry litter (CPL)], ASD2 (anaerobic soil disinfestation with 27.7 m³·ha⁻¹ of molasses, and 22 Mg·ha⁻¹ of CPL).

Table 3. Soil treatment effects on plant stem diameter, root weight and condition, root infesting root-knot juveniles (J2), and nonparasitic nematodes and nematode gall index of fresh-market tomato plants grown in Immokalee (University of Florida/Institute of Food and Agricultural Science/Southwest Florida Research and Education Center) and Citra (University of Florida/Plant Science Research and Education Unit) in the spring of 2015.^z

Treatments ^y	Stem diam (mm)	Root wt ^x (g)	Root condition ^w (0–5 scale)	Root-knot nematodes (J2/g root)	Nonparasitic nematodes (number/g root)	Gall index ^v (0–10 scale)
Immokalee, FL						
CSF	21.26	48.29	2.93 a	2.05	11.56	1.13 a
ASD1	20.31	45.06	2.05 b	0.56	7.71	0.63 b
ASD2	19.07	45.36	2.46 ab	1.02	9.96	1.26 a
<i>P</i> value	0.10	0.65	0.03	0.12	0.40	0.01
Citra, FL						
CSF	15.08 b	29.56	2.46	10.59	11.20	5.48 a
ASD1	20.57 a	36.56	2.63	5.45	14.32	3.52 b
ASD2	22.17 a	33.15	2.74	13.24	12.68	5.05 ab
<i>P</i> value	0.01	0.79	0.52	0.31	0.88	0.04

^zReported values are averages of four replications. Means followed by different letters within a column are significantly different at $P = 0.05$ by Duncan's multiple range test.

^yCSF (chemical soil fumigation, with Pic-Clor 60 at the rate of 224 kg·ha⁻¹ in Immokalee, and Paladin™ at the rate of 496 L·ha⁻¹ in Citra), ASD1 (anaerobic soil disinfestation with 13.9 m³·ha⁻¹ of molasses, and 22 Mg·ha⁻¹ of composted poultry litter), ASD2 (anaerobic soil disinfestation with 27.7 m³·ha⁻¹ of molasses, and 22 Mg·ha⁻¹ of composted poultry litter).

^xRoot weight on fresh weight basis.

^wRoot condition: 0 = clean, white roots, 5 = completely rotted and discolored roots.

^vNematode gall index: 0 = no galling, 10 = complete galling (Bridge and Page, 1980).

($P = 0.02$) effect was observed on the fruit firmness (Table 5). The fruit firmness was higher (lower deformation) in ASD1 and ASD2 than in CSF plots, which may be explained by the greater vigor and improved nutritional status of ASD plants as compared with CSF plants. Greater fruit firmness, without consequent negative effects on other commercial quality parameters (color, TSS, pH, DM) results in a longer shelf life (Meli et al., 2010) and is highly desirable considering that most of

the Florida tomato production is shipped to the North part of the United States. In Citra, none of the fruit quality parameters were significantly affected by soil disinfestation treatments (Table 5).

Soil treatment had a significant impact on the tomato fruit mineral content in Immokalee (Table 6). In Immokalee, K, Ca, and Mg content was significantly higher in fruit from ASD plots than from CSF. Total N was higher in fruit from CSF plots than in those

from ASD1 plots ($P = 0.01$), whereas treatment had no effect on the fruit P content. Among the micronutrients analyzed, fruit Fe content was significantly higher in CSF plots than in those subject to ASD2, and Mn fruit content was higher in CSF plots as compared with ASD fruits from both treatments, regardless of the molasses rate. No significant differences were observed for the content of B, Cu, Zn, Na, Mo, and Ni. In Citra, the fruit macro- and micronutrient content was not significantly influenced by soil treatments, except the case of Zn that was significantly higher ($P = 0.003$) in fruit from plots subjected to ASD1 and ASD2 than in those of CSF plots.

From these findings, we conclude that the application of ASD does not negatively affect the commercial tomato fruit quality, and that the quality and the mineral content of fruit produced with ASD is comparable or higher than that of fruit produced in CSF plots. Nevertheless, further studies should validate these results and consider the potential effects of ASD on other quality aspects, including the content of secondary metabolites that contribute to the health-promoting properties of tomatoes.

Conclusions

The results of this study, conducted on fresh-market tomato in two Florida locations (Immokalee and Citra), indicated that ASD, applied using a mixture of CPL and molasses as C source, may be potentially used as a sustainable alternative to conventional CSF for the control of plant-parasitic nematodes and weeds, without causing negative effects on tomato fruit yield and quality. The use of totally impermeable film, rather than transparent solarization mulch, did not hinder the development of anaerobic conditions, and avoided the need to substitute the solarization mulch with a second film. Although, cumulative redox potential was higher in ASD2 (27.7 m³·ha⁻¹ of molasses) as compared with ASD1 (13.9 m³·ha⁻¹ of molasses) plots, both treatments reached highly anaerobic conditions in both locations. In Immokalee, where the weed pressure was relatively low, ASD showed adequate herbicidal effect. However, in Citra, under relatively high nutsedge pressure, all treatments including the CSF resulted in an unacceptable level of nutsedge control. In both locations, the application of ASD resulted in root-knot nematode control similar to or greater than the CSF control. In Immokalee, total marketable yield was 26.7% and 19.7% higher in ASD2 and ASD1 plots than in CSF, respectively. While in Citra, regardless of the molasses rate, ASD provided a total marketable yield equivalent to the CSF. In both locations, the fruit quality was not influenced by soil treatments and was similar in ASD and CSF plots, except for the fruit firmness that in Immokalee was higher in ASD than in CSF plots. In terms of nutritional value, the macro- and micronutrient content of fruit produced in ASD plots was similar or higher than that of fruit produced in CSF plots.

Table 4. Soil treatment effects on tomato fruit size (scored by weight within each category) distribution from first, first and second harvest combined, and season total harvest (three harvests combined) in Immokalee (University of Florida/Institute of Food and Agricultural Science/Southwest Florida Research and Education Center) and in Citra (University of Florida/Plant Science Research and Education Unit) in the spring of 2015.^z

Treatments	First harvest (Mg·ha ⁻¹)				First and second harvest (Mg·ha ⁻¹)				Total season harvest (Mg·ha ⁻¹)				
	XL ^x	L	M	TMY	XL	L	M	TMY	XL	L	M	Cull	TMY
Immokalee, FL													
CSF ^y	15.4	4.6	0.0	20.0	28.2	7.3	0.3	35.9	33.3	10.5	5.2	4.9	49.0 b
ASD1	22.9	3.4	0.0	26.3	34.2	6.1	0.2	40.5	42.7	10.7	5.3	4.5	58.6 a
ASD2	19.4	3.9	0.0	23.2	34.0	7.4	0.4	41.9	43.4	12.6	6.1	4.3	62.1 a
<i>P</i> value	0.25	0.46	—	0.44	0.39	0.40	0.75	0.46	0.06	0.49	0.51	0.60	0.03
Citra, FL													
CSF	2.0	1.6	0.8	4.3	2.6	3.9	2.7	9.2	2.6	3.9	2.8	3.4	9.3
ASD1	2.8	2.6	0.7	6.1	4.1	5.1	3.5	12.7	4.1	5.2	4.1	4.9	13.4
ASD2	3.2	2.0	0.5	5.7	4.5	3.5	3.9	11.9	4.6	3.8	4.4	6.2	12.8
<i>P</i> value	0.49	0.31	0.71	0.47	0.16	0.41	0.17	0.30	0.14	0.45	0.13	0.11	0.19

^zReported values are averages of four replications. Means followed by different letters within a column are significantly different at *P* = 0.05 by Duncan's multiple range test.

^yCSF (chemical soil fumigation, with Pic-Clor 60 at the rate of 224 kg·ha⁻¹ in Immokalee, and Paladin™ at the rate of 496 L·ha⁻¹ in Citra), ASD1 [anaerobic soil disinfection with 13.9 m³·ha⁻¹ of molasses, and 22 Mg·ha⁻¹ of composted poultry litter (CPL)], ASD2 (anaerobic soil disinfection with 27.7 m³·ha⁻¹ of molasses, and 22 Mg·ha⁻¹ of CPL).

^xXL = extra-large (diameter (D) > 7.00 cm); L = large (6.35 < D < 7.00 cm); M = medium (5.72 < D < 6.43 cm); TMY = total marketable yield.

Table 5. Soil treatment effects on tomato fruit firmness (expressed as fruit deformation), skin color, Brix°, pH, and dry matter content at first harvest in Immokalee (University of Florida/Institute of Food and Agricultural Science/Southwest Florida Research and Education Center) and in Citra (University of Florida/Plant Science Research and Education Unit) in the spring of 2015.^z

Treatments	Deformation (mm)	Color stage (1–6 scale)	Brix°	pH	Dry matter (g·kg ⁻¹) ^x
Immokalee, FL					
CSF ^y	2.42 a	5.8	4.09	4.09	34.1
ASD1	2.01 b	5.6	4.08	4.12	32.7
ASD2	1.91 b	5.4	4.11	4.15	35.4
<i>P</i> value	0.02	0.17	0.93	0.42	0.40
Citra, FL					
CSF	3.79	5.5	5.35	4.41	61.5
ASD1	3.67	5.3	5.33	4.36	61.8
ASD2	3.81	5.6	5.55	4.45	61.7
<i>P</i> value	0.71	0.21	0.74	0.44	0.99

^zReported values are averages of four replications. Means followed by different letters within a column are significantly different at *P* = 0.05 by Duncan's multiple range test.

^yCSF (chemical soil fumigation, with Pic-Clor 60 at the rate of 224 kg·ha⁻¹ in Immokalee, and Paladin™ at the rate of 496 L·ha⁻¹ in Citra), ASD1 [anaerobic soil disinfection with 13.9 m³·ha⁻¹ of molasses, and 22 Mg·ha⁻¹ of composted poultry litter (CPL)], ASD2 (anaerobic soil disinfection with 27.7 m³·ha⁻¹ of molasses, and 22 Mg·ha⁻¹ of CPL).

^xDry matter is expressed in g·kg⁻¹ of fresh weight.

Table 6. Soil treatment effects on tomato fruit macro and micronutrient content at first harvest in Immokalee (University of Florida/Institute of Food and Agricultural Science/Southwest Florida Research and Education Center) and Citra (University of Florida/Plant Science Research and Education Unit) in the spring of 2015.^z

Element	Immokalee				Citra			
	CSF ^y	ASD1	ASD2	<i>P</i> value	CSF	ASD1	ASD2	<i>P</i> value
Macronutrient (g·kg ⁻¹ DW)								
N	30.2 a	25.9 b	28.0 ab	0.01	26.5	25.8	26.0	0.92
P	8.8	9.3	9.0	0.10	3.6	3.9	4.1	0.44
K	56.9 b	66.3 a	65.3 a	0.0001	35.4	39.0	40.6	0.49
Ca	2.4 b	2.8 a	2.7 a	0.002	2.0	2.2	1.9	0.51
Mg	2.6 b	2.8 a	2.8 a	0.02	1.7	1.8	1.7	0.73
Micronutrient (mg·kg ⁻¹ DW)								
Fe	46 a	43 ab	42 b	0.05	34	35	36	0.79
B	24	21	23	0.06	9	10	10	0.79
Cu	23	21	20	0.08	10	10	10	0.61
Zn	63	59	58	0.07	22 b	29 a	31 a	0.003
Mn	28 a	23 b	20 c	0.0001	14	14	16	0.59
Na	625	569	569	0.24	339	295	296	0.57
Mo	1.9	1.7	1.8	0.18	2.9	2.4	2.6	0.47
Ni	0.5	0.7	0.6	0.36	0.4	0.7	0.6	0.10

^zReported values are averages of four replications. For each location, means followed by different letters within a row are significantly different at *P* = 0.05 by Duncan's multiple range test.

^yCSF (chemical soil fumigation, with Pic-Clor 60 at the rate of 224 kg·ha⁻¹ in Immokalee, and Paladin™ at the rate of 496 L·ha⁻¹ in Citra), ASD1 [anaerobic soil disinfection with 13.9 m³·ha⁻¹ of molasses, and 22 Mg·ha⁻¹ of composted poultry litter (CPL)], ASD2 (anaerobic soil disinfection with 27.7 m³·ha⁻¹ of molasses, and 22 Mg·ha⁻¹ of CPL).

Overall, the results of the two locations demonstrate that the ASD technique may be a valid and sustainable alternative to the conventional CSF, and could be transferred at commercial level. As tested, molasses rates showed similar performance in terms of root-knot nematode and weed control, yield, and fruit quality; therefore, the lower molasses rate could be suggested to reduce the cost of the ASD treatment. However, further research is needed to validate the results of this study, consider other commercial crops, enhance the herbicidal activity, and test other potential C sources, to improve the ASD application technique for field production of vegetables and minimize the soil treatment costs.

Literature Cited

- Blok, W.J., J.G. Lamers, A.J. Termorshuizen, and G.J. Bollen. 2000. Control of soilborne plant pathogens by incorporating fresh organic amendments followed by tarping. *Phytopathology* 90: 253–259.
- Bonanomi, G., V. Antignani, C. Pane, and F. Scala. 2007. Suppression of soilborne fungal diseases with organic amendments. *J. Plant Pathol.* 89: 311–324.
- Bridge, J. and S.L.J. Page. 1980. Estimation of root-knot nematode infestation levels on roots using a rating chart. *Trop. Pest Mgt.* 26:296–298.
- Butler, D.M., N. Kokalis-Burelle, J.P. Albano, T.G. McCollum, J. Muramoto, C. Shennan, and E.N. Roskopf. 2014. Anaerobic soil disinfection (ASD) combined with soil solarization as a methyl bromide alternative: Vegetable crop performance and soil nutrient dynamics. *Plant Soil* 378:365–381.
- Butler, D.M., N. Kokalis-Burelle, J. Muramoto, C. Shennan, T.G. McCollum, and E.N. Roskopf. 2012a. Impact of anaerobic soil disinfection combined with soil solarization on plant-parasitic nematodes and introduced inoculum of soilborne plant pathogens in raised-bed vegetable production. *Crop Prot.* 39:33–40.
- Butler, D.M., E.N. Roskopf, N. Kokalis-Burelle, J.P. Albano, J. Muramoto, and C. Shennan. 2012b. Exploring warm-season cover crops as carbon sources for anaerobic soil disinfection (ASD). *Plant Soil* 355:149–165.
- Florida Department of Agriculture and Consumer Services (FDACS). 2005. Water quality/quantity

- best management practice manual for Florida vegetable and agronomic crops. 6 June 2015. <http://www.freshfromflorida.com/content/download/32110/789059/Bmp_VeggieAgroCrops2005.pdf>.
- Fiedler, S., M.J. Vepraskas, and J.L. Richardson. 2007. Soil redox potential: Importance, field measurements, and observations. *Adv. Agron.* 94:1–54.
- Fraisse, C.W., Z. Hu, and E.H. Simonne. 2010. Effect of El Niño–Southern oscillation on the number of leaching rain events in Florida and implications on nutrient management for tomato. *HortTechnology* 20:120–132.
- Freeman, J.H., E.J. McAvoy, N.S. Boyd, P.J. Dittmar, M. Ozores-Hampton, H.A. Smith, G.E. Vallad, and S.E. Webb. 2014. Tomato production, p. 183–204. In: G.E. Vallad, J.H. Freeman, and P.J. Dittmar (eds.). *Vegetable and small fruit production handbook of Florida, 2014–2015*. University of Florida Institute of Food and Agricultural Sciences, Gainesville, FL.
- Husson, O. 2013. Redox potential (Eh) and pH as drivers of soil/plant/microorganism systems: A transdisciplinary overview pointing to integrative opportunities for agronomy. *Plant Soil* 362: 389–417.
- Katase, M., C. Kubo, S. Ushio, E. Ootsuka, T. Takeuchi, and T. Mizukubo. 2009. Nematicidal activity of volatile fatty acids generated from wheat bran in reductive soil disinfection. *Nematological Research* 39:53–62.
- Kim, H.L., B.N. Jung, and B.K. Sohn. 2007. Production of weak acid by anaerobic fermentation of soil and antifungal effect. *J. Microbiol. Biotechnol.* 17:691–694.
- Lamers, J.G., W.T. Runia, L.P.G. Molendijk, and P.O. Bleeker. 2010. Perspectives of anaerobic soil disinfection. *Acta Hort.* 883:277–284.
- McCarty, D.G., S.E.E. Inwood, B.H. Ownley, C.E. Sams, A.L. Wszelaki, and D.M. Butler. 2014. Field evaluation of carbon sources for anaerobic soil disinfection in tomato and bell pepper production in Tennessee. *HortScience* 49: 272–280.
- Meli, V.S., S. Ghosh, T.N. Prabha, N. Chakraborty, S. Chakraborty, and A. Datta. 2010. Enhancement of fruit shelf life by suppressing N-glycan processing enzymes. *Proc. Natl. Acad. Sci. USA* 107:2413–2418.
- Meng, T., T. Zhu, J. Zhang, and Z. Cai. 2015. Effect of liming on sulfate transformation and sulfur gas emissions in degraded vegetable soil treated by reductive soil disinfection. *J. Environ. Sci.* 36:112–120.
- Momma, N. 2008. Biological soil disinfection (BSD) of soilborne pathogens and its possible mechanisms. *Jpn. Agr. Res. Q.* 42:7–12.
- Momma, N., Y. Kobara, and M. Momma. 2011. Fe²⁺ and Mn²⁺, potential agents to induce suppression of *Fusarium oxysporum* for biological soil disinfection. *J. Gen. Plant Pathol.* 77:331–335.
- Momma, N., K. Yamamoto, P. Simandi, and M. Shishido. 2006. Role of organic acids in the mechanisms of biological soil disinfection (BSD). *J. Gen. Plant Pathol.* 72:247–252.
- Mowlick, S., K. Hirota, T. Takehara, N. Kaku, K. Ueki, and A. Ueki. 2012. Development of anaerobic bacterial community consisted of diverse clostridial species during biological soil disinfection amended with plant biomass. *Soil Sci. Plant Nutr.* 58:273–287.
- Mowlick, S., T. Inoue, T. Takehara, N. Kaku, K. Ueki, and A. Ueki. 2013a. Changes and recovery of soil bacterial communities influenced by biological soil disinfection as compared with chloropicrin-treatment. *AMB Express* 3:46.
- Mowlick, S., T. Takehara, N. Kaku, K. Ueki, and A. Ueki. 2013b. Proliferation of diversified clostridial species during biological soil disinfection incorporated with plant biomass under various conditions. *Appl. Microbiol. Biotechnol.* 97:8365–8379.
- Noling, J.W., J.P. Gilreath, and D.A. Botts. 2006. Alternatives to methyl bromide soil fumigation for Florida vegetable production, p. 121–126. In: S.M. Olson and D.N. Maynard (eds.). *Vegetable production handbook for Florida, 2006–2007*. University of Florida Institute of Food and Agricultural Sciences, Gainesville, FL.
- Oka, Y. 2010. Mechanisms of nematode suppression by organic soil amendments—A review. *Appl. Soil Ecol.* 44:101–115.
- Ozores-Hampton, M., F. Di Gioia, S. Sato, E. Simonne, and K. Morgan. 2015. Effects of nitrogen rates on nitrogen, phosphorous, and potassium partitioning, accumulation, and use efficiency in seepage-irrigated fresh market tomatoes. *HortScience* 50:1636–1643.
- Ozores-Hampton, M., P.A. Stansly, and T.P. Salame. 2011. Soil chemical, physical, and biological properties of a sandy soil subjected to long-term organic amendments. *J. Sustain. Agr.* 35:243–259.
- Ozores-Hampton, M., E. Simonne, F. Roka, K. Morgan, S. Sargent, C. Snodgrass, and E. McAvoy. 2012. Nitrogen rates effects on the yield, nutritional status, fruit quality, and profitability of tomato grown in the spring with subsurface irrigation. *HortScience* 47:1129–1133.
- Rabenhorst, M.C. and K.L. Castenson. 2005. Temperature effects on iron reduction in a hydric soil. *Soil Sci.* 170:734–742.
- Rich, J.A., Jr., R. Brito, R. Kaur, and J.A. Ferrell. 2009. Weed species as hosts of Meloidiogyne: A review. *Nematropica* 39:157–185.
- Roskopf, E., P. Serrano-Pérez, J. Hong, U. Shrestha, M.C. Rodríguez-Molina, K. Martin, N. Kokalis-Burelle, C. Shennan, J. Muramoto, and D. Butler. 2015. Anaerobic soil disinfection and soil borne pest management, p. 277–305. In: Meghvansi and Varma (eds.). *Organic amendments and plant disease control*. Springer International Publishing.
- Sargent, S.A., J.K. Brecht, Q. Wang, and T. Olczyk. 2005. Handling Florida tomatoes—round and roma types. *Univ. Florida, Inst. Food Agr. Sci., Electronic Data Info. Source, SS-VEC-928*. 29 June 2015. <<http://edis.ifas.ufl.edu/pdf/files/VH/VH07900.pdf>>.
- Shennan, C., J. Muramoto, J. Lamers, M. Mazzola, E.N. Roskopf, N. Kokalis-Burelle, N. Momma, D.M. Butler, and Y. Kobara. 2014. Anaerobic soil disinfection for soil borne disease control in strawberry and vegetable systems: Current knowledge and future directions. *Acta Hort.* 1044:165–175.
- Shinmura, A., N. Sakamoto, and H. Abe. 1999. Control of *Fusarium* root rot of Welsh onion by soil reduction (abstract in Japanese). *Jpn J Phytopathol* 65:352–353.
- Simonne, E.H. and G.J. Hochmuth. 2011. Soil and fertilizer management for vegetable production in Florida, p. 3–16. In: S.M. Olson and S. Bielinsky (eds.). *Vegetable production handbook for Florida, 2011–2012*. University of Florida Institute of Food and Agricultural Sciences, Gainesville, FL.
- Smajstrla, A.G. and R. Muñoz-Carpena. 2011. Simple water level indicator for seepage irrigation. *Univ. Florida, IFAS, EDIS publ. AE085*. 6 June 2015. <<http://edis.t.ifas.ufl.edu/pdf/files/AE/AE08500.pdf>>.
- Stall, W.M. and J.P. Morales-Payan. 2003. The critical period of nutcase interference in tomato. *Southwest Florida Research and Education Center: University of Florida*.
- Strauss, S.L. and D.A. Kluepfel. 2015. Anaerobic soil disinfection: A chemical-independent approach to pre-plant control of plant pathogens. *J. Integr. Agr.* 14:2309–2318.
- Thoden, T.C., G.W. Korhals, and A.J. Termorshuizen. 2011. Organic amendments and their influences on plant-parasitic and free-living nematodes: A promising method for nematode management? *Nematology* 13(2):133–153.
- USDA Natural Resources Conservation Service (USDA-NRCS). 2010. Field indicators of hydric soils in the United States, a guide for identifying and delineating hydric soils. In: L.M. Vasilas, G.W. Hurt, and C.V. Noble (eds.). *National Technical Committee for Hydric Soils*.
- USDA National Agricultural Statistics Service (USDA-NASS). 2015. Vegetable 2014 summary. U.S. Dept. Agr., Washington, DC. 18 Aug. 2015. <<http://usda.mannlib.cornell.edu/usda/current/VegeSumm/VegeSumm-01-29-2015.pdf>>.
- U.S. Department of Agriculture (USDA), Agriculture Marketing Service. 1997. United States standards for grade of fresh tomatoes. Washington, DC. 18 Aug. 2015. <http://www.ams.usda.gov/sites/default/files/media/Tomato_Standard%5B1%5D.pdf>.
- U.S. Environmental Protection Agency. 1997. Test methods for evaluating solid waste, physical/chemical methods: EPA Publ. SW-846. Microwave assisted acid digestion of siliceous and organically based matrices. Method 3052, Office of Solid Waste, USEPA, Washington.
- van Agtmaal, M., G.J. van Os, W.G. Hol, M.P.J. Hundscheid, W.T. Runia, C.A. Hordijk, and W. de Boer. 2015. Legacy effects of anaerobic soil disinfection on soil bacterial community composition and production of pathogen-suppressing volatiles. *Front. Microbiol.* 6:1–12.
- Zotarelli, L., J.M. Scholberg, M.D. Dukes, R. Munoz-Carpena, and J. Icerman. 2009. Tomato yield, biomass accumulation, root distribution and irrigation water use efficiency on a sandy soil, as affected by nitrogen rate and irrigation scheduling. *Agr. Water Mgt.* 96:23–34.