Research Article

Rebuilding Soil Ecosystems for Improved Productivity in Biosolarized Soils

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Disinfecting soil can reduce or eliminate crop loss from soilborne pathogens, parasitic nematodes, and weed competition. Biosolarization combines biotoxic products from organic matter decomposition and heat from solarization. While biosolarization offers an organic option for soil pest control and avoids human and environmental health risks associated with chemical fumigants, it still has broad negative impacts on microbial communities. Quickly reestablishing these communities can be key in preventing resurgence in disease pressure and in maximizing nutrient use efficiency. The objective of this study was to determine the ability of fertilization source, N fertilization rate, and/or inoculate to rebuild an active soil ecosystem in biosolarized soils by measuring nematode community structure, microbial biomass, and C and N mineralization in soil as well as kale yield and quality. The study was conducted using potted kale grown in biosolarized soils. Treatments were bare soil, receiving no fertilization, and soils receiving two different rates of organic, composted broiler litter or mineral fertilizer. Half of the pots in each treatment received a locally sourced microbial inoculant (LEM). Among the nonfertilized treatments, the soils that received applications of LEM mineralized more nitrogen and produced higher yields. Soils that received the highest rate of compost immobilized the greatest proportion of nitrogen applied, were the most cold-tolerant, and produced the largest yields. None of the kale that received heavy mineral fertilization without LEM inoculation survived a hard freeze; however, the ones that received LEM applications were able to partially recover. We also found that kale grown with lower rates of N (50 kg·ha⁻¹), applied as compost plus LEM, had the greatest magnesium leaf content and were more economically efficient while producing an equally abundant and nutritious food.

1. Introduction

Soil disinfestation has been recognized and used as an important mechanism since the beginning of the 20th century for controlling the more obscure pests operating below ground [1]. Employing some form of soil disinfestation can be highly effective in reducing or eliminating crop loss from soilborne pathogens, plant-parasitic nematodes, and weed competition. Heat, flooding, and fumigation are all strategies used to disinfest soil [1]. For over 70 years, methyl bromide (MB) fumigation was the leading form of soil disinfestation, until the Montreal Protocol and the U.S. Clean Air Act mandated a phase-out of its import and

manufacture in 2001 [2]. The impact of soil disinfestation on the productivity of agriculture in the U.S. or the world is not small. Approximately 48,000 metric tons of MB is used globally each year in agriculture [3]. Also, it is estimated that eliminating MB as a soil fumigant option may cost U.S. producers and consumers over \$1 billion a year [4]. While there are other soil fumigant chemicals available, such as chloropicrin and metam sodium, there is a great deal of research being done to find safer, more sustainable alternatives including cultural practices, host resistance and stimulation of plant growth, and control of disease through the addition of plant growth-promoting rhizobium (PGPR) [5–10]. Solarization is a milder treatment than most

chemical treatments. By trapping the sun's energy as heat under transparent, polyethylene plastic covering, solarization heats up the soil enough to weaken or kill plant-parasitic organisms in the soil. Biosolarization is the incorporation of easily decomposable organic matter, irrigation, and tarping with polyethylene mulch [10]. This creates a combination of biotoxic products from the decomposition of the organic matter and heat created from the solarization and has been proven to be comparable to MB in its effectiveness in killing fungal pathogens as well as plant-parasitic nematodes [10]. While biosolarization offers an organic option for soil pest control and avoids the human and environmental health risks associated with chemical fumigants, it still creates an issue of having a broad negative effect on the soil microbial community. If the world is going to move towards agricultural systems that focus on maintaining and improving soil health, there needs to be work done to find ways of controlling potentially devastating disease and pest pressure without also eliminating the numerous beneficial services provided to the soil by nonpathogenic soil microbial community.

Healthy soils provide sustained biological productivity, promote environmental quality, and maintain plant and animal health [11]. According to the USDA NRCS Soil Health Initiative, in order to truly care for the health of our nation's soils so that they can provide these important ecosystem services, they must be treated as living ecosystems teeming with billions of bacteria, fungi, and other microbes.

One important ecosystem service that a healthy, functioning soil offers agricultural producers is nutrient cycling. The microbial community within soil is responsible for transforming nutrients into a plant-available form from organic and inorganic sources, for maintaining nutrients in the soil, and for the release of nutrients near roots where the plants can access them [12, 13]. Considering these two vitally important services provided by a rich and balanced soil ecosystem, it is easy to understand the importance of quickly reestablishing a variety of microbes into disinfested soils. One way of doing this is adding organic amendments such as composts or manures. Organic material offers a source of labile carbon and associated micro- and macronutrients which feed the soil biome and organic material can serve as an inoculant introducing an abundance of active soil fauna and biota. Another way of helping to ensure the delivery of services from healthy soil biomes is through inoculation with specific soil microbes such as arbuscular mycorrhizal fungi [14, 15], or mixtures of specific plant growth-promoting rhizobacteria, including species of *Pseudomonas*, *Azotobacter*, and *Bacillus* [16]. Inoculating soils with microbial communities, thriving in healthy local forest systems can help ensure multifunctional edaphon likely to flourish. These inoculant amendments can be used in addition to, or instead of organic material amendments. One such amendment is Local Effective Microorganisms (LEM).

LEM is a solution of microbes that can be homemade by producers at a low cost. LEM is based on the commercial product called effective microorganisms or EM™, which is a

mixed culture of beneficial and naturally occurring microorganisms that was developed by Japanese scientist Dr. Teruo Higa. The microorganisms in EM™ include species of photosynthetic bacteria (*Rhodospseudomonas palustris* and *Rhodobacter sphaeroides*), lactobacilli (*Lactobacillus plantarum*, *L. casei*, and *Streptococcus lactis*), yeasts (*Saccharomyces* spp.), and actinomycetes (*Streptomyces* spp.) [17]. According to the EM Research Organization (<http://www.emrojapan.com/how/>), the purpose of these beneficial organisms is to improve crop growth and yield by increasing photosynthesis, producing bioactive substances such as hormones and enzymes, controlling soil diseases, and accelerating the decomposition of lignin materials in the soil. In parts of Latin America and Asia, Dr. Higa's theory of EM™ has led to the development of a locally produced version of these effective microorganisms (LEMs). The microbes found in LEM are derived from yeast, raw milk, and decomposing leaf litter in forested areas on or near the same farm to which the LEM is to be applied. The aim of this research is to explore the effect LEM has on improving soil health of soils that have been disinfested using biosolarization through the reestablishment of a rich microbial population.

Microbial activity is assessed by analyzing microbial biomass, CO₂ respiration, and nematode assemblage structure [18, 19]. Being one or two steps higher in the food chain than bacteria and fungi, nematodes serve as integrators of physical, chemical, and biological properties related to their food resources [20]. Nematodes occupy nearly every niche in the soil food web and can, therefore, reveal a great deal about changes in soil microbial structure as a whole. By counting and identifying nematodes extracted from our soils, we can use various indices to determine the condition and function of the soil ecosystems they are a part of [21–25].

The objectives of this study were to assess the impact of LEM inoculant and fertilization source and N fertilization rate on soils' ability to rebuild a healthy microbial community after biosolarization. The impact of each of these treatments was determined by analyzing (a) size and activity of microbial populations by comparing measurements of soil microbial biomass carbon, CO₂-flush carbon, and nitrogen mineralization; (b) soil food web complexity and structure by comparing nematode trophic group and community structure indices; and (c) crop performance by comparing yield, chlorophyll level, and elemental nutrient concentrations in kale leaves.

2. Materials and Methods

2.1. Experimental Design. Soils for this study were biosolarized at the J. Phil Campbell Research and Education Center in Watkinsville, Georgia (33°52'N, 83°27'W). The soil at the site is a fine kaolinitic, thermic Typic Kanhapludults and had been managed under certified organic management since 2012. It is in a Cecil sandy loam series with a 2–6 percent slope [26].

In order to biosolarize the soils before treatment application, annual ryegrass (*Lolium perenne* L.spp.

FIGURE 1: Soil biosolarization temperatures at a depth of 10 cm, from 9 May 2017 to 21 July 2017.

multiflorum (Lam.)) was tilled into 7 m strips that were then irrigated and covered with clear, polyethylene plastic. Biosolarization was started in early May. Temperature under the plastic was monitored at a depth of 10 cm to make sure temperatures of 35–40°C were maintained for at least 4–6 weeks (Figure 1). Once temperatures began to drop in late July, the top 10 cm of soil was collected, thoroughly mixed, and used to fill the one hundred pots used for the treatment replications. After pots were filled with biosolarized soil, treatments were applied and kale (*Brassica oleracea*) cv “Toscano” was direct seeded into the pots. The kale was later thinned to a density of three plants per pot. The potted kale was grown outdoors, hand-watered with chlorine-free water, and covered during rain events to prevent saturation and leaching.

In this experiment, there were three types of variables tested which were combined, resulting in a total of ten sets of treatments. The variables were (1) fertilizer source (composted, organic broiler litter versus mineral fertilizer) (2) rate of nitrogen applied ($134.5 \text{ kg-N}\cdot\text{ha}^{-1}$ vs. $50.4 \text{ kg-N}\cdot\text{ha}^{-1}$ vs. $0 \text{ kg-N}\cdot\text{ha}^{-1}$) (3) inoculation (receiving microbial inoculant versus not receiving microbial inoculant). The broiler litter used was made from locally sourced, organic broiler litter. The broiler litter treatments either received applications of LEM at turning throughout the composting process or were produced using only water at turning. The mineral fertilizer used was a combination of ammonium sulfate, triple superphosphate, and potassium chloride. The mineral fertilizer was mixed to match the proportions of nitrogen, phosphorus, and potassium determined from analysis of the composted broiler litter used. The LEM inoculant was applied at a rate of $18,337 \text{ L}\cdot\text{ha}^{-1}$. LEM is made by cultivating the O horizon biome from a local, undisturbed forest floor with a growing media under anaerobic conditions. After six weeks, the solid LEM was extracted in a 1:16 sugar:water solution. Additional details on producing the LEM inoculant can be found in supplementary materials. There were ten replications of each of the treatment combinations (listed in Table 1).

2.2. Sampling and Analysis of Soil Microbial Biomass Carbon and CO_2 -Flush Respiration. Soils were sampled for soil microbial biomass carbon (SMBC) and CO_2 -flush analysis at the time of harvest. After kale plants were harvested from the pots, the soil was removed to a depth of 15 cm and mixed thoroughly. Approximately 300 g of the mixed soil was collected and stored at 4°C until analysis. SMBC was determined using the fumigation extraction methods described by [27]. CO_2 -flush carbon was determined according to the methods described by [28].

2.3. Sampling and Analysis of Nitrogen Mineralization. Soil samples were taken for nitrogen (N) mineralization incubations immediately after fertilizer and inoculant application. KCl extractions were performed on samples before incubation (Day 0) and again after 7 and 28 days of incubation. Soils were sampled to a depth of 15 cm and stored at 4°C before being extracted. Subsamples (5 g) were extracted with a 2 M KCl solution according to [29]. NH_4^+ and NO_3^- were measured from the extracts colorimetrically on a Tecan Infinite M200 Pro nanoquant using the vanadium (III) reduction method and the salicylic method, respectively.

Because two rates of N were applied, we calculated and presented the mineralized N as the proportion of N applied, $\text{MinN}/\text{AppliedN}$ where $\text{MinN}/\text{AppliedN} = \text{Day Xi} - \text{Day 0}/\text{AppliedN}$. The $\text{MinN}/\text{AppliedN}$ was calculated for each treatment to compare the amount of nitrogen mineralized by the soil microbial communities relative to how much was added to each system. The exceptions to this are the B and LB treatments. Because these treatments received no N fertilization, the MinN is presented without dividing by the N applied.

2.4. Sampling and Analysis of Nematode Community Structure. Samples for nematode community analysis were taken eight weeks after application. Soil cores were taken from 0–10 cm [30–34] and then were kept refrigerated at 4°C before extraction. One hundred cubic centimeters of soil were extracted at room temperature (20°C) using the centrifugal flotation method as described by [35]. Extracts were kept refrigerated (4°C) before the nematode populations were counted. Extracts were brought up to 40 mL, gently and thoroughly shaken, before a 10 mL aliquot was pipetted into a dish for counting. Nematodes were counted live, on an inverted, compound microscope and identified to trophic group or family according to [36, 37]. Nematodes were analyzed to family and assigned colonizer-persister (c-p) values according to [38]. Maturity Index (MI), Structural Index (SI), and Enrichment Index (EI) values were calculated according to [39]. These calculations can be found in the supplementary materials.

2.5. Analysis of Kale Quality, Yield, and Frost Tolerance. Twice during the growing season, a CCM-200 Chlorophyll Content Meter (Opti-Sciences, Hudson, New Hampshire,

TABLE 1: Treatment labeling and abbreviations.

Treatment name and abbreviation	Composted broiler litter vs. mineral fertilizer	High vs. low rate of fertilizer	LEM vs. no LEM
LEM mineral—high (LM135)	Mineral fertilizer	134.5 kg·N·ha ⁻¹	LEM
Control mineral—high (M135)	Mineral fertilizer	134.5 kg·N·ha ⁻¹	None
LEM compost—high (LC135)	Composted broiler litter	134.5 kg·N·ha ⁻¹	LEM
Control compost—high (C135)	Composted broiler litter	134.5 kg·N·ha ⁻¹	None
LEM mineral—low (LM50)	Mineral fertilizer	50.4 kg·N·ha ⁻¹	LEM
Control mineral—low (M50)	Mineral fertilizer	50.4 kg·N·ha ⁻¹	None
LEM compost—low (LC50)	Composted broiler litter	50.4 kg·N·ha ⁻¹	LEM
Control compost—low (C50)	Composted broiler litter	50.4 kg·N·ha ⁻¹	None
LEM bare (LB)	None	None	LEM
Control bare (B)	None	None	None

USA) was used to collect nondestructive chlorophyll level in kale leaves. Leaf, stem, and root weights were taken at harvest. Roots were double rinsed and patted dry before being weighed. All portions of the harvested kale were then dried at 60°C for 48 hours. Stabilized dry weights were rerecorded, and the dried leaves were ground to pass a 2 mm sieve using a Wiley mill (Thomas Scientific, Swedesboro, New Jersey USA) and then to pass through a 1-mm sieve using a Cyclotec sample mill (FOSS, Eden Prairie, Minnesota USA) for sample analysis. The ground samples were then digested using 20% HCl [40]. The digestants were analyzed for Ca, Mg, and Zn using an A Analyst 200 Atomic Absorption Spectrometer (Perkin Elmer, Waltham, Massachusetts, USA).

There was an unexpected hard frost that severely damaged the kale plants. The extent to which they were damaged and the extent to which they recovered varied greatly between treatments. The percent of plants that survived the freeze was recorded and is reported as survivability.

3. Results and Discussion

3.1. Soil Microbial Biomass Carbon and CO₂-Flush Carbon. There were no significant differences in SMBC between soils treated with LEM and soils not treated with LEM (Figure 2). Soil that received compost at the greater rate, with and without LEM had significantly greater SMBC than soil that received mineral fertilizer at the lower rate with and without LEM and soil that received mineral fertilizer at the greater rate without LEM. Soils that received compost at the greater rate also had the lowest amounts microbial activity measured as CO₂ flush. The incongruency between these sets of data highlights the fact that SMBC and CO₂ respiration, while both a measurement of the microbial community in the soil, represent different properties of these communities. SMBC is helpful in understanding the number of nutrients and elements that are being held in the bacterial and fungal pool [41]. It cannot be determined by SMBC, however, which fractions of this biomass are dead, dormant, or active and is therefore, not an accurate indicator of biological activity in the soil [42]. Measuring CO₂, on the other hand, assesses the respiration of soil microbes. This provides a proxy for the rate of mineralization and decomposition by a soil's microbial community and is likely why CO₂-flush values have been found to correlate with N mineralization [28].

Comparing Figures 2(a) and 2(b), the SMBC and CO₂-flush values from the different treatments have an inverse relationship. This suggests that treatments with high CO₂ values and low SMBC values have a more rapid rate of turnover/mineralization of carbon and that this carbon is likely being mineralized from the labile SMBC pool in the soil, while the treatments with low CO₂-flush values are mineralizing less carbon from the soil and maintaining more in the microbial biomass pool.

3.2. Nitrogen Mineralization. By assessing the amount of nitrogen mineralized from the time that treatments were applied, as a proportion of the nitrogen that was applied (MinN/AppliedN), we are able to compare the nitrogen-cycling potential of the organisms within each treatment. This calculation allows for assessment of soil microbial provisioning of plant-available nitrogen. Positive values indicate net mineralization of nitrogen, and negative values indicate net immobilization of nitrogen by the soil microbial communities. N mineralization values for the soils that did not receive N fertilization (LB and B) were compared separately from the other soils because they could not be represented as proportions of N applied. Between these soils, significantly more nitrogen was mineralized in the LB soils than in the B soils at day 7 of incubation (Figure 3). Since there was no evidence of increased general microbial activity, measured by either SMBC or CO₂ respiration, in the LB soils versus the B soils (Figure 2), we speculate that LEM treatment increased the proportion of nitrogen-provisioning microbes compared to the bare soils that did not receive LEM (Mahmud et al., in review). There were no significant differences in N mineralization between fertilization treatments at days 7 or 28 of incubation. The greatest proportion of N was immobilized in LC135 soils followed by C135 soils. The lowest CO₂-flush values and greatest SMBC values were also measured in these two treatments. The N immobilization data supports the assertion that the increased CO₂-flush values are linked to reduced microbial biomass pools.

3.3. Nematode Community Structure. After application of all treatments, the populations of bacterial-feeding nematodes increased compared to biosolarized soils (post), except for LM135 (Figure 4). Treatments: LC50, LC135,

FIGURE 2: Left: soil microbial biomass (SMBC). Right: CO₂ flush. Treatments applied to biosolarized soils were bare control with no amendment (B), bar with LEM inoculant (LB), compost at 50 kg·N·ha⁻¹ and at 135 N·ha⁻¹ with and without LEM (C50, C135, LC50, and LC135), respectively, and mineral fertilizer at 50 kg·N·ha⁻¹ and at 135 N·ha⁻¹ with and without LEM (M50, M135, LM50, and LM135). Different letters indicate significant differences ($\alpha = 0.10$).

FIGURE 3: Treatments: bare control with no amendment (B), bare with LEM inoculant (LB), compost at 50 kg·N·ha⁻¹ and at 135 N·ha⁻¹ with and without LEM (C50, C135, LC50, and LC135), respectively, and mineral fertilizer at 50 kg·N·ha⁻¹ and at 135 N·ha⁻¹ with and without LEM (M50, M135, LM50, and LM135). B and LB treatments must be analyzed separately. While they have had day 0 nitrogen subtracted from their mineralized nitrogen values, because there was no N added to the pots, they have not been divided by any amount and are therefore on a much larger scale than the other treatments. Different lowercase letters indicate significant differences at day 7 of incubation. Different capital letters (A&B for amended treatments; Q&R for bare treatments) indicate significant differences at day 28 of incubation ($\alpha = 0.10$).

C135, LM50, and LM135 showed a significant increase in fungal-feeding nematode populations after treatment applications. Within the high-rate mineral fertilizer treatments, LM135 had significantly greater fungal-feeding

nematodes than M135. There were no significant increases from population levels post-solarization in plant-parasitic nematode populations or in Tylenchidae among any treatments.

FIGURE 4: The x axis includes baseline soils before and after biosolarization (Pre and Post) and soils that received treatments applied to biosolarized soils: bare control with no amendment (B), bare with LEM inoculant (LB), compost at 50 kg·N·ha⁻¹ and at 135 N·ha⁻¹ with and without LEM (C50, C135, LC50, and LC135), respectively, and mineral fertilizer at 50 kg·N·ha⁻¹ and at 135 N·ha⁻¹ with and without LEM (M50, M135, LM50, and LM135). Different letters indicate significant differences ($\alpha = 0.10$).

As was expected, the LB and B treatments, which received no amendments, had the lowest EI values and the LC135 and C135, which had the higher rate of compost applied to them, and had the highest values (Table 2). The LC50 treatments also had significantly higher EI values than the C50 treatments. The EI values of the LM135 and M135 soils were lower than LC135 and C135 soils, respectively, and the LM50 and M50 values were lower than those of the LC50

and C50, respectively. This is likely because the compost amendments, while equivalent to the mineral amendments in N, P, and K, contain large amounts of organic matter and are naturally rich in detritivores.

The EI is calculated based on the numbers of nematodes that quickly take advantage of nutrient inputs (enrichment). These advantageous nematodes are also the ones that have low c-p values. Therefore, when they increase in a

TABLE 2: Nematode community index values.

Treatment	Maturity index (MI)	Structural index (SI)	Enrichment index (EI)
B	2.35 ab	61.80 a	5.62 e
LB	2.27 ab	55.57 ab	9.83 de
C50	2.19 abc	52.20 ab	16.87 cd
LC50	2.05 cd	53.13 ab	36.69 ab
C135	1.97 de	40.45 bc	32.67 ab
LC135	1.82 e	10.23 d	39.09 a
M50	2.27 ab	54.63 ab	11.87 de
LM50	2.17 bc	39.53 bc	18.11 cd
M135	2.02 cd	21.5 cd	19.28 cd
LM135	2.28 ab	44.43 abc	27.57 bc
Std. Error	0.06	6.46	4.70

Treatments applied to biosolarized soils: bare control with no amendment and no LEM inoculant (B), bare with LEM inoculant (LB), compost at 50 kg-N-ha⁻¹ and at 135 N-ha⁻¹ with and without LEM (C50, C135, LC50, and LC135), respectively, and mineral fertilizer at 50 kg-N-ha⁻¹ and at 135 N-ha⁻¹ with and without LEM (M50, M135, LM50, and LM135). Different letters indicate significant differences within columns ($\alpha = 0.10$).

TABLE 3: Kale yield and quality measurements.

Treatment	Chlorophyll ₁ 12/01/17	Chlorophyll ₂ 12/20/17	Leaf yield (g)	Leaf yield/N applied	Survival (%)
B	46 c	71 abc	50 d	—	80
LB	47 c	69 abc	81 c	—	70
C50	55 ab	77 ab	105 bc	107 ab	87
LC50	57 ab	75 ab	126 b	128 a	70
C135	63 a	80 a	166 a	63 cd	90
LC135	62 a	79 a	177 a	67 cd	100
M50	47 c	67 c	54 cd	55 cd	38
LM50	50 bc	67 bc	69 cd	70 cd	27
M135	44 cd	61 cd	—	—	0
LM135	43 d	63 d	36 cd	14 e	22
Std. error	3.4	4.4	13.5	11.3	—

Treatments applied to biosolarized soils: bare control with no amendment and no LEM inoculant (B), bare with LEM inoculant (LB), compost at 50 kg-N-ha⁻¹ and at 135 N-ha⁻¹ with and without LEM (C50, C135, LC50, and LC135), respectively, and mineral fertilizer at 50 kg-N-ha⁻¹ and at 135 N-ha⁻¹ with and without LEM (M50, M135, LM50, LM135). Different letters indicate significant differences within columns ($\alpha = 0.10$). Leaf Yield/N applied reported as g dried leaves g⁻¹ N applied to each pot. Survival indicates the % of plants that recuperated after a hard freeze in January 2018.

population, the MI and SI tend to decrease resulting in an inverse relationship between the indices. Considering the given EI values, it is logical that the LC135 and C 135 treatments have correspondingly low MI and SI values, and LB and B have correspondingly high MI and SI values. M135 has both low EI values but also low MI and SI values. This suggests low maturity and structure in M135 nematode communities despite there not being a large increase in low c-p value, detritivore-feeding nematodes.

3.4. Kale Yield and Quality. Kale grown in soils that received the compost had the highest chlorophyll values, the greatest yield per pot, and the best frost survival (Table 3). Soils that received no compost, only mineral fertilizers had much lower frost survival. Treatment M135 had no plants that recuperated after a hard freeze while the LM135 plants had a few. Most of the kale in the B and LB treatments survived the freeze but had lower chlorophyll content and leaf yield than compost-treated kale. When considering the kale yield in proportion with the amount of N applied to each treatment, LC50 and C50 faired best among the amended soils. Between

the two bare treatments, LB had a greater yield than treatment B. It is not clear why the kale plants that received mineral amendments were more susceptible to frost damage than either the compost or the nonamended treatments.

The nitrogen concentration in the kale leaves was similar between amended treatments; however, the kale leaves from treatment LB had a significantly higher concentration of nitrogen than the B treatments (Table 4). For clarity, leaf nutrient concentrations are for 100 gm of leaf material and not based on yield. Leaf Mg concentration was greatest for LC50 as was leaf yield per N applied. Bare treatment had the least K concentration in kale leaves and the least N concentration, while one of the greatest leaf N concentrations were in the LB leaves. LM135 had among the highest concentrations for Ca, Mg, and Zn. An excess of one of the minerals like K could have had an antagonistic effect on the kale's ability to take up adequate Mg or Ca. Deficiencies in these minerals would have caused the plants to be less tolerant to stress [43] but upon performing elemental analysis of the leaf tissue, elemental concentrations in the mineral treatments were similar to other treatments.

TABLE 4: Elemental concentrations of kale leaves at time of harvest.

Treatment	K (mg/100 g)	Ca (mg/100 g)	Mg (mg/100 g)	Zn (mg/100 g)	N (mg/kg)
B	1175 d	196 ab	35.5 bc	3.07 b	910 c
LB	1209 cd	199 a	35.6 bc	3.14 b	1371 ab
C50	1265 abc	198 a	35.7 bc	3.35 b	1384 ab
LC50	1297 ab	191 abc	36.3 a	3.30 b	1118 bc
C135	1275 ab	183 bc	35.1 c	3.28 b	1209 abc
LC135	1307 a	177 c	35.4 bc	3.06 b	1248 abc
M50	1257 abc	188 abc	35.3 bc	3.10 b	1491 ab
LM50	1237 bcd	198 ab	35.5 bc	3.02 b	1551 a
M135	—	—	—	—	—
LM135	1264 abc	209 a	35.9 ab	3.90 a	1089 abc
Std. error	81.2	6.85	0.25	0.16	117.0

Treatments applied to biosolarized soils: bare control with no amendment and no LEM inoculant (B), bare with LEM inoculant (LB), compost at 50 kg-N-ha⁻¹ and at 135 N-ha⁻¹ with and without LEM (C50, C135, LC50, and LC135), respectively, and mineral fertilizer at 50 kg-N-ha⁻¹ and at 135 N-ha⁻¹ with and without LEM (M50, M135, LM50, and LM135). Different letters indicate significant differences within columns ($\alpha=0.1$).

4. Conclusion

Biosolarization significantly diminished plant-parasitic nematode populations which did not rebound during the subsequent growing season. Most trophic groups from the free-living nematode populations, however, were able to recuperate to at least prebiosolarization numbers. The performance of kale grown in biosolarized soils was promoted by compost application which increased both yield and cold tolerance. The LC50 treatment produced the most yield per unit of nitrogen applied, the greatest leaf concentration of Mg. Soils amended with LEM combined with the lower rate of compost did not differ with treatments considered to be greatest in K, or Ca. These results indicate that kale grown with lower rates of N (50 kg-ha⁻¹), applied as compost, are more economically efficient while producing an equally abundant and nutritious food. LEM significantly improved soil nitrogen mineralization, kale leaf yield, and leaf nitrogen content in soils that did not receive any compost amendment or mineral fertilizer. Though it is unclear why kale that received mineral fertilizer had low cold tolerance. Plants that received a high rate of mineral fertilization and no LEM application had zero percent survival from frost damage while LEM inoculated counterparts had a twenty-two percent survival. In conclusion, we would highly recommend the addition of compost to biosolarized soils to improve yield and hardness of crops and would also recommend the application of LEM, especially in the absence of compost amendments.

Data Availability

The spreadsheet data used to support the findings of this study may be released upon application to Dorcas H. Franklin, who can be contacted at dfrankln@uga.edu.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

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Supplementary Materials

Making LEM and FALSE—LEM, maturity index, structural index, and enrichment index. (*Supplementary Materials*)

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