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The Application of Cyanobacteria as a Biofertilizer for Okra (*Abelmoschus esculentus*) Production with a Focus on Environmental and Ecological Sustainability

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Abstract: Cyanobacteria, an important addition to biofertilizers, are gaining popularity for their multifaceted benefits in sustainable agriculture and ecosystem restoration. However, harmful algal blooms (HABs) in freshwater, predominantly caused by cyanobacteria, prevent sunlight penetration into the water and develop hypoxic and anoxic conditions. We collected cyanobacteria slurry from Lake Jesup (Central Florida, USA), repurposed it as a biofertilizer, and incorporated it in a typical South Florida calcite soil for high-value okra (*Abelmoschus esculentus*; var: Clemson spineless) production. Experiments were conducted at the Organic Garden Shade House and Greenhouse located inside the main campus of the Florida International University (FIU), FL, USA. A two-year experiment with four different treatments was conducted, namely, (a) control (C; no fertilizer applied), (b) total synthetic (TS), (c) total biofertilizer (TB; only cyanobacteria biofertilizer was applied), and (d) half and half (HH; 50% biofertilizer + 50% synthetic fertilizer), which were arranged in a randomized complete block design (RCBD) with six replications for each treatment. Our results indicate that TB and TS produced about 29 to 33% higher SPAD (soil plant analytical development) readings than the control. The absence of interveinal chlorosis (yellowing of leaves) in the TB and HH treatments suggests that the cyanobacteria-based biofertilizer had a role in supplying one of the critical micronutrients, iron (Fe). Analysis of the biofertilizer indicated 2000 ppm Fe content, which directly supports our observation. Similarly, average plant height (61 cm), yield (130 gm per pot), and crop biomass (67 gm) productions were significantly higher in TB than in the control. Overall, this study documents the potential of cyanobacteria biofertilizers as a viable option compared to synthetic fertilizers for sustainable crop production and soil health improvement.

Keywords: cyanobacteria; biofertilizer; okra; organic nutrient source; Florida; sustainable agriculture

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1. Introduction

Biofertilizers are organic substances containing abundant bio-stimulants and microorganisms. Biofertilizers include algae, plant growth-promoting rhizobacteria (PGPR), and other mineralizing microorganisms, all of which can potentially contribute to mobilizing nutrients in the soil, thereby enhancing soil and crop productivity. Additionally, biofertilizers are considered organic substitutes for synthetic chemical fertilizers to promote organic farming practices. Cyanobacteria (prokaryotic microorganisms), a group of photosynthesizing bacteria, are gaining acceptance as organic biofertilizers due to their multifaceted benefits to soil health and crop production. Unlike other symbiotic microorganisms, cyanobacteria do not depend on a host for their growth and development and have a nitrogen-fixing capability of 25 to 60 kg N ha^{-1} in the soil [1]. Some of the benefits of cyanobacteria biofertilizers are (a) the release of extracellular polymeric substances (polysaccharides) to improve soil aggregate stability, the (b) release of plant growth-promoting hormones [2], (c) the increase in leaf chlorophyll content and antioxidant enzyme activity, and (d) maintaining a symbiotic relationship with other microorganisms and enhancing microbial biomass C in soil [3]. The biofilm layer (from biological soil crusts; BSCs) formed from the extracellular polysaccharides of cyanobacteria serves as a habitat for other organisms in the soil. This synergistic relationship often improves soil biodiversity and ecosystem resiliency. Cyanobacteria are also capable of sequestering carbon in their cells (like C_4 plants) [4].

However, the presence of cyanobacteria in freshwater ecosystems causes a major threat by creating harmful algal blooms (HABs). HABs prevent sunlight penetration in water and create eutrophic conditions. Once the algal blooms complete their life cycle and decompose, secondary microbes decompose the biomass, resulting in oxygen depletion. In addition to creating hypoxic conditions in waterbodies, cyanobacteria produce cyanotoxins, which are harmful to humans, animals, and aquatic organisms. Specifically, favorable weather conditions (warm temperature and high relative humidity) in Florida have led to high HABs in major lakes, generally covering 60–80% of the lake area during peak algal bloom season (May to mid-September). For the last couple of decades, the occurrence of harmful algal blooms has been a recurrent problem in Florida, causing environmental and ecological degradation and economic loss. One of the remedial measures is the unwanted cyanobacteria slurry collected from HABs disposed of as a landfill material. In this study, we repurposed the harvested cyanobacteria slurry to produce biofertilizers for growing high-value organic vegetables. Specifically, we used cyanobacteria biofertilizers for okra (specialty crop) cultivation. Okra is one of the most important vegetable crops grown in Florida, contributing a significant revenue to the state economy. Okra is grown approximately 1000 to 1500 acres annually in Miami-Dade County, South Florida [5]. Okra is also a short-duration crop (generally takes 65 to 70 days from planting to harvesting) and prefers hot weather, which makes it easier to grow okra during the summer months in South Florida.

The application of cyanobacteria as a biofertilizer in row crops, such as rice, wheat, soybean, cotton, and maize, has been well documented [6,7]. However, most of these studies have been conducted either directly in the field (such as a paddy rice field) or used cyanobacteria inoculants for their experiments. Studies focusing on the application of cyanobacteria as a biofertilizer (after drying and processing) for producing crops and maintaining soil quality are still underexplored. Furthermore, there is a lack of understanding of the beneficial effects of cyanobacteria as a biofertilizer on crop production and soil health in the United States. Typically, chicken manure, turkey manure, pig/cow manure, vermicompost, and other composting materials are used for organic farming in the USA. To the best of our knowledge, no experiments conducted in the USA have explored the effect of naturally occurring cyanobacteria as a biofertilizer for crop production and soil health improvement. Therefore, our research objective was to evaluate the performance of cyanobacteria biofertilizers in the geographical and climatic conditions (sub-tropical) of South Florida. We also expect that our study will improve the basic knowledge about the numerous benefits that this biofertilizer can offer in an agricultural production system. The specific objectives of this study were 1) to evaluate the effects of cyanobacteria biofertilizers on plant growth and physiological parameters of okra and 2) to assess the efficacy of cyanobacteria biofertilizers for improving soil properties compared to synthetic fertilizers.

2. Materials and Methods

2.1. Cyanobacteria Collection and Biofertilizer Processing

Cyanobacteria slurry materials were collected from Lake Jesup (28°43′ N, 81°13′ W) located in Central Florida, USA. The slurry was collected from different locations of the Lake. The cyanobacterial slurry was collected in five-gallon buckets for easy transportation and brought back to the University campus for further processing. Large plant roots and other visible unwanted things were discarded carefully from the collected materials. The collected slurry was air dried (after spreading on large plastic trays) in the greenhouse for 3 to 4 weeks to obtain dried cyanobacteria materials (Figure 1). The slurry on the plastic trays was stirred up occasionally to expedite the drying process. Dried cyanobacteria were then powdered using a mechanical grinder and stored in colored plastic bags for future applications. Storing cyanobacteria biofertilizers in colored plastic bags were reported to increase shelf life of the biofertilizer [8]. Cyanobacteria can also be used in the soil as semi-solid wet materials; however, it is difficult to measure the nutrient concentrations in wet cyanobacteria, and the wet materials need to be applied immediately after collection. Dried and powdered cyanobacteria materials were mixed with dried soil before filling up the pots for a uniform distribution of the biofertilizer. A recent study reported that dry cyanobacteria material works more effectively as fertilizer than wet materials for crop production [9]. The concentrated slurry had about 72 to 75% water (roughly 12 to 15% dry matter content). The dry matter content of the slurry was processed (after drying) to produce the biofertilizer.

2.2. Experimental Design and Sample Collection

This two-year experiment was conducted in the greenhouse (year one) and in the shade house (year two) at Florida International University (FIU), Florida, USA. Soils (Biscayne marly silt loam) used for this experiment were collected from a farmer's field located in Homestead, FL. The physicochemical properties of the background soil are presented in Table 1. Okra (*Abelmoschus esculentus*; var: Clemson spineless) was grown in three-gallon (11.4 L) plastic nursery pots spaced 2.5 ft (0.91 m) apart from each other (Figure 1D). Four experimental treatments, namely, the (a) control (C; no fertilizer applied), (b) total synthetic (TS; synthetic urea 46-0-0 and sulfate of potash 0-0-51 were applied), (c) total biofertilizer (TB; only cyanobacteria biofertilizer was applied), and (d) half and half (HH; 50% biofertilizer + 50% synthetic fertilizer were applied), were assigned in a randomized complete block design (RCBD) with six replications for each treatment for both year one and two experiments. Recommended doses of N and K for okra production are 135 kg N ha^{-1} and 112 kg K_2O ha^{-1}. The amount of biofertilizer needed for each treatment (TB and HH) was calculated based on the nutrient contents of the dried cyanobacteria biofertilizer (Table 2). Composite soil samples (0 to 15 cm depth) collected from the field for these two experiments were analyzed for physicochemical properties. It can be noted that the collected soil was an actual representation of typical South Florida organic soils, which have alkaline limestone parent material (high $CaCO_3$). Large roots and stones were removed from the soil through sieving before filling the pots.

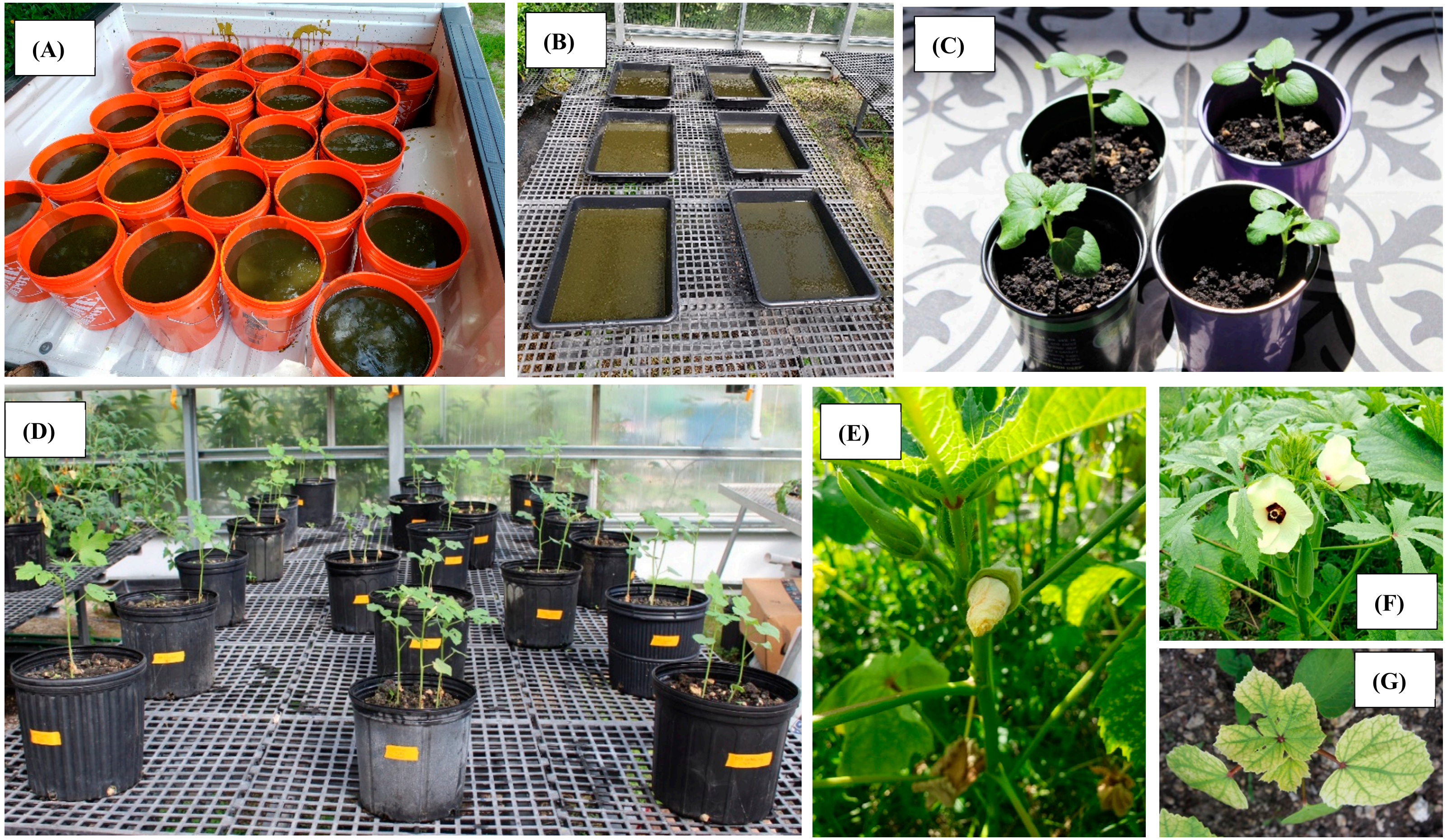

Figure 1. Pictures of okra growth in greenhouse settings. (**A**) Cyanobacteria slurries collected in five-gallon buckets, (**B**) drying of cyanobacteria materials, (**C**) one-week-old okra seedlings, (**D**) greenhouse settings of the experiment (including all treatments), (**E**) okra flowers, (**F**) okra flower and fruits together, and (**G**) early nutrient deficiency in control plants.

Table 1. Physicochemical properties of the soil used for the experiments.

Parameters	Unit	Value
pH		8.01
Total C (TC)	%	6.07 ± 0.42
Total N (TN)	%	0.34 ± 0.06
Total P (TP) *	ppm	103 ± 27
Potassium (K)	ppm	175 ± 42
Calcium (Ca)	ppm	13,604 ± 2722
Magnesium (Mg)	ppm	255 ± 29
Sulphur (S)	ppm	79.20 ± 8.24
Zinc (Zn)	ppm	24.48 ± 5.69
Copper (Cu)	ppm	32.80 ± 4.91
Sodium (Na)	ppm	51.73 ± 11.73

* Analyzed using the Solórzano and Sharp (1980) method and the EPA method 365.1. Analyzed through Mehlich 3 extraction.

Table 2. Physicochemical properties of the dried and powdered cyanobacteria biofertilizer.

Parameters	Unit	Value
Total C (TC)	%	19.56 ± 0.71
Total N (TN)	%	1.79 ± 0.07
Total P (TP)	%	0.02 ± 0.00
Total S (TS)	%	0.13 ± 0.01
Potassium (K)	%	0.06 ± 0.01
Calcium (Ca)	%	6.12 ± 0.70
Magnesium (Mg)	%	0.12 ± 0.01
Iron (Fe)	ppm	2005 ± 160
Manganese (Mn)	ppm	132.56 ± 13.35
Zinc (Zn)	ppm	53.34 ± 0.69
Copper (Cu)	ppm	29.51 ± 4.80
Boron (B)	ppm	95.37 ± 4.21
Molybdenum (Mo)	ppm	1.82 ± 1.01
Nickel (Ni)	ppm	4.34 ± 1.78

Three okra seeds were planted for each pot, and thinning was performed at the four-leaf stage to keep the healthiest plant in the pot. Soils from each pot were also collected at harvesting and analyzed for physicochemical properties.

2.3. Plant Height, Stem Diameter, Leaf Chlorophyll Content, Crop Biomass, and Okra Fruit Yield

Plant height (cm), stem diameter (cm), and leaf chlorophyll content of the okra plant from each pot were recorded at four, five, six, and seven weeks after planting and at harvesting. The experiment was terminated at 67 days after sowing (DAS), and fruit yield was obtained by weighing the harvested okra from each pot. Plant height and stem diameter (five readings from each plant) were measured using meter sticks and slide calipers, respectively. The average leaf chlorophyll content of the developed upper leaves was recorded using the Soil-Plant Analyses Development (SPAD) 502 Plus Chlorophyll Meter (Konica Minolta, Inc, Ramsey, NJ, USA). Fresh shoot and root samples were collected during harvesting (67 DAS). Shoot samples (stem and leaves) were taken from the first node up. Roots were carefully washed to remove soil particles. Both shoot and root samples were dried at 60 °C for 72 h to obtain dry aboveground and belowground biomasses.

2.4. Laboratory Analyses

Soil pH was measured (1:1 dried ground soil to water ratio) using a seven excellence pH meter (Mettler Toledo, Columbus, OH, USA). Analysis of total nitrogen and total carbon content of the collected dried ground cyanobacteria, soil, and plant samples were performed using a CN analyzer (LECO company, St. Joseph, MI, USA) following the Dumas dry combustion method. The dried ground cyanobacteria and plant samples were digested using nitric acid–hydrogen peroxide to determine micro- and macronutrient contents. Later, the digested and filtered samples were analyzed using Perkin Elmer Optima 8300 ICP-OES (Waltham, MA, USA). Dried ground soil samples were extracted using the Mehlich 3 extraction procedure. The filtered samples were analyzed using Perkin Elmer Optima 8300 ICP-OES.

2.5. Statistical Analyses

All data in this study are presented as mean and standard deviation. Statistical analyses were performed using SAS 9.4 (SAS Institute NC, Cary, NC, USA). The treatment effects on different plant growth parameters, soils, and plant physicochemical properties were evaluated using a one-way analysis of variance (ANOVA). A mean comparison was performed using Tukey–Kramer and Fisher's LSD post hoc tests at $p < 0.10$ and $p < 0.05$. Differences at mean values $p < 0.05$ were considered statistically significant, while differences at mean values $p < 0.05$ were discussed.

3. Results and Discussion

3.1. Physicochemical Properties of the Biofertilizer

The N content in the cyanobacteria biofertilizer ranged from 1.56 to 1.94% (Table 2), which appears to be higher compared to the N content reported in a couple of published articles [10,11]. However, most of those previous studies produced cyanobacteria under controlled conditions (tunnel house, small microcosm) or cultured in a rice paddy field. We collected the free-flowing cyanobacteria slurries (naturally grown) from Lake Jesup in Central Florida. The lake often receives high amounts of nutrients as runoff from the adjacent agricultural areas [12], which resulted in higher nutrient contents in our cyanobacteria dry mix. Cyanobacteria adsorb or fix atmospheric N and store it in their vacuoles, but the magnitude of N fixation commonly depends on the cyanobacteria species. For instance, *Anabaena* and *Nostoc* cyanobacteria species used for biofertilizer preparation (when inoculated in a controlled environment) have higher N content than other species [1,13]. We collected free-flowing cyanobacteria from the lake where *Anabaena*, *Nostoc*, and *Clostridium* species were commonly present. It was difficult to segregate those species from the slurry mixture; therefore, it was expected that our samples had a mixture of different species of cyanobacteria.

The C:N ratio of our biofertilizer was in the range of 12:1 to 14:1, which is lower than organic wastes, like straw 100:1, sawdust 200:1, leaves, and plant stalks. Therefore, it takes months to process the organic wastes, such as straw and sawdust (to lower the C:N ratio), before application in the field. So, it was a major advantage of our newly developed biofertilizer where a lower C:N ratio can potentially influence the net N mineralization in the soil and thus, the biofertilizer was ready to be applied immediately in the soil as a source of plant nutrients.

Fe (>2000 ppm) and Mg (>1200 ppm) were in higher concentrations in the biofertilizer produced in this study (Table 2). South Florida soils are mostly porous sandy loam, with alkaline soil pH and low organic matter content. Thus, growers in South Florida (specifically organic growers) often face production issues due to Fe deficiency in the soil (personal communication with the growers), resulting from calcareous parent materials. Therefore, it is expected that the cyanobacteria biofertilizer developed in this study will be able to reduce the Fe deficiency problem in South Florida soils. More about the effects of Fe and Mg on okra production at different treatments are discussed later in this manuscript (Section 3.2(a)).

3.2. Plant Physiological Parameters

(a) Leaf chlorophyll content—SPAD values

Chlorophyll content in plant leaves is an indicator of leaf N content, chloroplast development, and overall plant health. The soil–plant analytical development (SPAD) meter is a simple, rapid, and non-destructive instrument used to analyze leaf chlorophyll contents of plants. SPAD generates three-digit readings (unitless), where higher SPAD values indicate high leaf N content and a healthy plant. We started collecting SPAD data four weeks after planting the crop when the leaves were old enough to produce SPAD readings. SPAD readings were recorded every week afterwards (until week 7) and at harvesting to obtain the leaf chlorophyll content of okra (Figure 2). In this study, SPAD values were highly variable throughout the experiment; however, prominent differences among treatments were observed at five and six weeks after planting the crop. Average SPAD values of TS and TB pots were 1.29 and 1.33 times higher, respectively (significant at $p < 0.05$), than control pots, indicating that plants in TS and TB pots had higher plant vigor and leaf chlorophyll content than pots that received no fertilizer treatment. A study conducted by Romanowska-Duda et al. [14] documented similar observations and found that cyanobacteria biofertilization increased leaf chlorophyll content, photosynthetic productivity, and crucial plant growth enzymes.

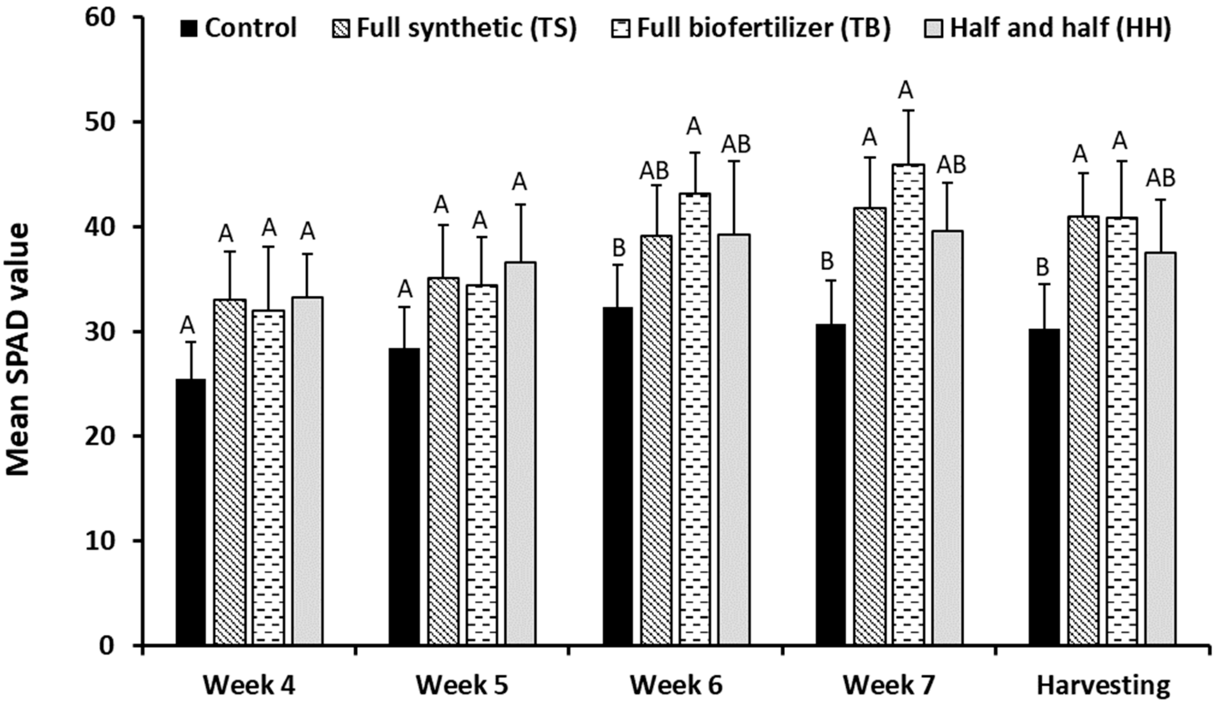

Figure 2. Soil–plant analytical development (SPAD) values of okra at different growth stages. Similar uppercase letters indicate no significant difference at $p < 0.10$.

SPAD values of control pots (average 29.44 for all five data points) started declining after week 6 (32.31), with a parallel observation of leaf yellowing in some pots Common leaf yellowing (chlorosis) of plants is an indication of nutrient (N, S, Fe, Mn) deficiency, depending on the location of the deficiency symptoms (older vs. younger leaves) and type of the chlorosis (full chlorosis or interveinal). However, a prominent pale-yellow leaf color (interveinal chlorosis) and stunted plant growth were observed 50 days after planting (mostly in younger leaves), indicating the possibility of Fe deficiency in those plants. Visual observation of the chlorosis is displayed in Figure 3. A sign of the yellowing of leaves was also observed in TS plants, but the number of leaves affected was fewer than in control plants. South Florida soils are deficient in Fe (due to alkaline soil pH), and deficiency symptoms are common among plants that received no fertilizer treatments (control pots). Comparing the treatments, no yellowing of leaves was observed in TB and HH pots, resulting from high Fe content (more than 2000 ppm) in the biofertilizer prepared from cyanobacteria (Table 2). The cyanobacteria biofertilizer was reported to provide a significant amount of Fe in the soil along with other nutrients [9]. Additionally, Mg, which is a main component of chlorophyll, was in abundant quantity (1200 ppm) in the cyanobacteria biofertilizer prepared for this experiment. To support our observation, we analyzed the plant samples from each pot after harvesting., Plant analysis data indicate that Fe content was significantly higher in TB (258 ppm) and HH (220 ppm) than in the control (152 ppm), further confirming the fact that chlorosis in the control plants was Fe deficiency symptoms.

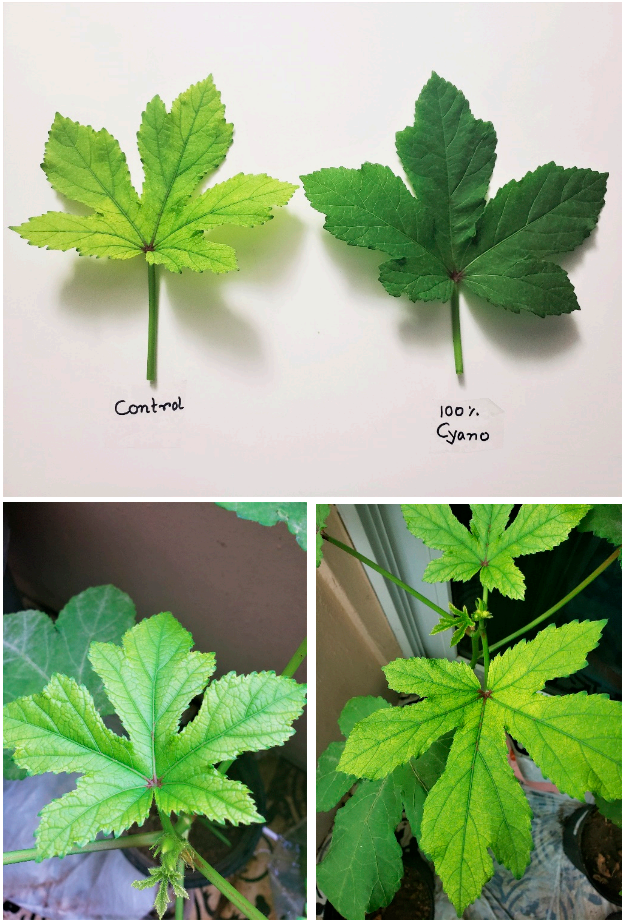

Figure 3. Visual representation of possible Fe deficiency (interveinal chlorosis) in control plants from the shade house study. The top picture shows the difference between leaves from full cyanobacteria pots (green color) and control pots (light green color with pale yellow spots).

Average SPAD values for TS pots at weeks four and five were 2 to 4% higher than TB; however, starting from week 6, we observed higher SPAD values in TB pots than in TS pots. A common explanation is the synthetic fertilizers, specifically urea (producing ammonia after hydrolysis), which tend to be easily volatized from the soil profile [15], and thus, less amount of soil N was available for plants at five weeks after planting. A lower amount of N in the leaf produced lower SPAD values for TS pots.

The average SPAD value combining all treatments (excluding the control) at harvesting was 7% lower than the value at week seven, confirming that leaf senescence close to harvest would have resulted in a lower SPAD value. It should be noted that the SPAD value depends on various factors, including leaf age, environmental conditions, and other factors [16]. It is noted that lower SPAD values close to the crop harvesting stage were observed in previous studies [17,18].

(b) Plant height, stem diameter, and crop biomass production and okra yield

Average plant height, stem diameter, and crop biomass (shoot and root) are summarized in Table 3. Plant heights of TS (62.8 cm) and TB (61.3 cm) were similar to each other (not significantly different); however, both TS and TB had significantly higher plant heights than the control treatment. Plant height is an active indicator of vegetative growth or the vigor of the plant. The difference in plant height of okra under various treatments during weeks five and seven is easily visible in Figure 4.

Table 3. Physiological parameters (plant height, stem diameter, aboveground and belowground biomasses) and okra yield under different treatments. Experiments were conducted at the FIU Organic Garden Shade House and the FIU greenhouse. Data presented are an average of year one and year two experiments.

Treatments	Plant Height *	Stem Diameter *	Shoot Dry Weight¶	Root Dry Weight *	Shoot/Root	Yield¶
	cm	cm	g			g/pot
Control	45.3 ± 3.68 b	0.78 ± 0.08 a	26.71 ± 4.61 b	5.61 ± 0.88 b	4.69	73.38 ± 5.27 b
TS	62.8 ± 4.81 a	1.08 ± 0.16 a	47.92 ± 6.99 ab	8.37 ± 1.95 ab	6.14	120.98 ± 9.42 a
TB	61.3 ± 5.16 a	1.11 ± 0.13 a	55.01 ± 7.62 a	11.58 ± 2.16 a	3.78	130.34 ± 8.78 a
HH	58.3 ± 3.68 ab	1.05 ± 0.13 a	37.07 ± 3.92 ab	7.59 ± 0.78 ab	4.21	110.18 ± 12.36 ab

TS = only (100%) synthetic fertilizer; TB = only (100%) cyanobacteria biofertilizer; HH = half (50%) synthetic fertilizer and half (50%) cyanobacteria biofertilizer. * Similar lowercase letters indicate no significant difference at $p < 0.10$; ¶ similar lowercase letters indicate no significant difference at $p < 0.05$.

Stem diameter, which is also an indicator of plant water status [19], was highest in okra grown with TB (average 1.11 cm) followed by TS, HH, and the control. It is often described that a higher stem diameter indicates better plant growth [20] and can result in high crop biomass production. Consequently, a similar trend was observed for plant biomass production, where total biomass production (shoot and root combined) in TB and TS was 1.60 to 1.80 times higher than the control. An interesting observation was that the higher root biomass in TB pots (average 11.6 gm) was higher than in other treatments. The shoot to root ratio was the lowest in TB pots (3.78), indicating that the cyanobacteria biofertilizer was able to improve root structure more than shoot biomass production in this study. Biofertilizer pot (TB) soils at harvesting had numerically higher (196 ppm) total P content than synthetic pots (TS; 117 ppm), which was another possible reason for better root growth in plants that received biofertilizer treatments. A research study documented that cyanobacteria are capable of increasing root length, root volume, and quality for vegetable crops [21], which is a major advantage of those crops for efficient nutrient uptake from the soil.

Yield analysis of okra plants from four treatments is presented in Table 3. The average yields (gm per pot) of TB (130) and TS (121) were significantly higher ($p < 0.001$) than the control. Biofertilizers act as natural sources of plant nutrients and promote overall plant health and productivity. An interesting observation during these two experiments was quicker physiological developments (specifically plant height and stem diameter) of the plants that received biofertilizer treatments (both in TB and HH) than other treatments. Specifically, flowering and fruit settings in biofertilizer-treated pots were observed at least 7 to 10 days earlier than other treatments. We did not measure plant growth regulators and hormones in the biofertilizer soils, but previous studies reported that cyanobacteria exudates are capable of adding bioactive compounds and growth regulators in the soil [2,22], which improve plant physiological parameters.

Figure 4. Visual representation of okra plants (shade house study) received different treatments (**A**) at week 4 and (**B**) at week 6 after planting. Treatments in this picture are as follows (from left to right): full synthetic fertilizer (TS), full cyanobacteria biofertilizer (TB), HH (half (50%) synthetic fertilizer and half (50%) cyanobacteria biofertilizer), and control. Pictures are from the shade house study.

3.3. Soil Properties at Harvesting

Soils collected after harvesting were analyzed for physicochemical properties (Table 4). A major observation was lower soil pH in biofertilizer-treated pots (both in TB and HH) than in other pots. The decomposition of cyanobacteria often releases organic acids in the soil during decomposition [2], which explains the slightly acidic pH in the TB soils at harvesting. Since South Florida soils are alkaline (with high $CaCO_3$ contents), cyanobacteria biofertilizer decomposition will help reduce the soil pH and solubilize the nutrients. Even for a shorter study (for about 70 days), the amount of C (10.77%) and N (0.53%) contents in soils at TB pots at harvesting were significantly higher than the control (C, 5.46% and N, 0.30%). Mg, which is a major component of chlorophyll, was also significantly higher in TB pots than other treatments. Phosphorus contents (ppm) in the soil at harvesting from TB (196) and HH (141) pots were significantly higher ($p < 0.05$) than control pots possibly because of the ability of the biofertilizer to enhance P bioavailability in the soil through solubilization. Overall, most of the nutrients were higher in TB soils at harvesting compared to the control, an indication of improved soil nutrient status in cyanobacteria-applied pots.

In a study conducted in Nigeria, Agwa et al. [23] found that cyanobacteria inoculants significantly increased soil nutrient status for okra production.

Table 4. Okra leaves' nutrient content at harvest. Data presented are an average of year one and year two experiments.

Treatments	C	N	P	K	Ca	Mg	S	Fe	Mn	B
	%							ppm		
Control	27 ± 6 b	1.9 ± 0.4 b	0.18 ± 0.04 b	1.3 ± 0.41 a	2.9 ± 0.7 b	0.62 ± 0.18 a	0.32 ± 0.14 a	152 ± 27 c	17 ± 4 b	48 ± 11 a
TS	36 ± 11 ab	2.9 ± 0.6 ab	0.26 ± 0.07 ab	2.0 ± 0.62 a	4.1 ± 1.1 ab	0.85 ± 0.29 a	0.33 ± 0.14 a	173 ± 42 bc	37 ± 8 a	40 ± 9 a
TB	44 ± 7 a	3.7 ± 0.5 a	0.32 ± 0.07 a	2.3 ± 0.59 a	4.8 ± 1.0 a	0.95 ± 0.21 a	0.35 ± 0.17 a	258 ± 31 a	28 ± 11 ab	42 ± 13 a
HH	38 ± 10 ab	3.1 ± 0.6 a	0.22 ± 0.07 ab	1.7 ± 0.24 a	4.5 ± 0.9 ab	0.88 ± 0.27 a	0.30 ± 0.16 a	220 ± 29 ab	33 ± 7 a	51 ± 7 a

TS = only (100%) synthetic fertilizer; TB = only (100%) cyanobacteria biofertilizer; HH = half (50%) synthetic fertilizer and half (50%) cyanobacteria biofertilizer; C, N, P, K, Ca, Mg, and S are presented as %, and Fe, Mn, and B are presented as ppm. Similar lowercase letters indicate no significant difference at $p < 0.10$.

3.4. Environmental Implications

A major component of this research study was to foster environmental and ecological sustainability. We removed free-flowing cyanobacteria from the Lake, which is otherwise a great threat to the aquatic life due to eutrophication (reducing the dissolved oxygen content of the water). Additionally, a cyanobacteria slurry in the lake causes a nuisance (foul smell) to the adjacent neighborhood. Repurposing cyanobacteria as a biofertilizer in field-scale agricultural settings presents an opportunity to explore their potential benefits for crop production and soil quality improvement in diverse agroecosystems. The balanced array of plant nutrients, including both macro- and micronutrients, provided by the cyanobacteria biofertilizer (Table 2) offers an alternative to synthetic fertilizers for crop production in South Florida and beyond. As a result, it is expected to reduce nutrient runoff to the surface water systems and improve overall environmental quality. As mentioned earlier, South Florida soils are predominantly porous sandy loam, alkaline, and low in organic matter (OM) content. Our biofertilizer contributes organic carbon to the soil (OC content in cyanobacteria soil was two times higher than control soil; Table 5), which is expected to improve soil quality and aggregate stability. Better soil structure not only increases nutrient retention and availability to plants but also reduces nutrient leaching into freshwater systems, such as Lake Jesup and surrounding areas, further supporting environmental and ecological sustainability efforts.

Table 5. Soil nutrient content after okra harvesting. Data presented are an average of year one and year two experiments.

Treatments	pH	C	N	P	K	Ca	Mg	S	Na
		%		ppm					
Control	7.69	5.46 ± 0.49 b	0.296 ± 0.05 c	86 ± 19 b	154 ± 28 b	12,064 ± 1223 b	242 ± 37 b	37 ± 6 b	47 ± 15 a
TS	7.84	8.44 ± 1.27 a	0.438 ± 0.07 ab	117 ± 39 ab	388 ± 111 a	13,289 ± 1361 b	354 ± 38 a	102 ± 18 a	67 ± 14 a
TB	6.63	10.77 ± 1.53 a	0.533 ± 0.06 a	196 ± 41 a	301 ± 120 ab	20,850 ± 2209 a	384 ± 29 a	89 ± 11 a	82 ± 24 a
HH	7.89	9.15 ± 1.11 a	0.407 ± 0.05 b	141 ± 22 a	256 ± 62 a	13,927 ± 1621 b	325 ± 41 a	116 ± 22 a	74 ± 22 a

TS = only (100%) synthetic fertilizer; TB = only (100%) cyanobacteria biofertilizer; HH = half (50%) synthetic fertilizer and half (50%) cyanobacteria biofertilizer; C and N are presented as %, and all other nutrients are presented as ppm. About 3.42 and 1.71 gm of dried biofertilizer were added to 1 kg of soil for TB and HH treatments, respectively. Similar lowercase letters indicate no significant difference at $p < 0.10$.

On an economic aspect, we calculated that the application of plant nutrients through cyanobacteria biofertilizers will reduce the cost of production by 12 to 15% compared to using synthetic fertilizers. In addition, the use of cyanobacteria biofertilizers can fully or partially substitute chemical fertilizers without any loss of crop yield (okra). Therefore, cyanobacteria biofertilizers can help growers generate high-value organic okra and stay competitive in the market. South Florida soils are deficient in Fe (due to alkaline parent materials), and organic growers often face serious Fe deficiency challenges during crop production. Most of the growers apply chelated Fe as an organic Fe source, which substantially increases the cost of production. Cyanobacteria biofertilizers can provide 2000 ppm of Fe in the soil (Table 2), and substituting expensive organic Fe sources with cyanobacteria biofertilizers can significantly alleviate production costs for these growers.

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