Variety Trials

Yield and Fruit Quality Traits of Two Plantain Cultivars Grown at Two Locations in Puerto Rico under Black Leaf Streak Disease Pressure

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SUMMARY. Plantain (*Musa balbisiana* AAB) is a tropical rhizomatous perennial plant in the genus *Musa* spp., closely related to banana (*Musa acuminata* AAA). It is an important cash crop and staple for inhabitants in many parts of the world, including various ethnic groups in the United States. Black leaf streak disease (BLSD) or black sigatoka, caused by *Pseudocercospora fijiensis* (formerly *Mycosphaerella fijiensis*), is responsible for significant losses of this crop due to the high susceptibility of the most economically important cultivars. BLSD does not immediately kill plantain plants, but it causes severe leaf necrosis, which results in reduced photosynthetic area, thereby adversely impacting bunch weight and fruit production. Without cultural and chemical control, yields can be reduced by 20% to 80%, depending on severity. This study evaluated a BLSD-resistant cultivar, FHIA-21, against Maricongo, a standard commercial cultivar with no BLSD tolerance, at two locations in Puerto Rico on Ultisol (Corozal site) and Oxisol (Isabela site) soils. Total number of fruit and bunch yield were significantly higher at Isabela, with BLSD severity being significantly lower at this location. Average fruit production of 'FHIA-21' was significantly higher than that of 'Maricongo' at both locations, with fruit yields of 122,522 and 99,948 fruit/ha at Isabela and Corozal, respectively. Overall, fruit of 'FHIA-21' were significantly longer and had greater diameters than those of 'Maricongo'. At Isabela, the mean bunch fruit weight was significantly higher for 'FHIA-21', but both cultivars exceeded the minimum local marketable fruit weight criterion of 270 g. At both locations, the numbers of functional leaves present at flowering and at harvest were significantly higher for 'FHIA-21' than for 'Maricongo', indicating more availability of photosynthetic area for 'FHIA-21' during the fruit-filling period. There were no significant differences between cultivars regarding the concentration of starch and soluble sugars for green fruit. Regarding ripe fruit, 'FHIA-21' had a significantly higher concentration of soluble sugars and less starch. In this study, 'FHIA-21' had good resistance against BLSD and, if accepted by consumers, is a viable alternative to current commercial cultivars. We also conclude from this study that the expression of the *Banana streak virus* (BSV) in planting material of this cultivar remains an unknown threat in yield decline of 'FHIA-21'.

Plantain is a tropical rhizomatous perennial plant closely related to banana, and it is an important staple food crop in Africa, Central America, South America,

Oceania, southeast Asia, and the Caribbean Basin (Robinson and Galán-Saúco, 2010). Most cultivated polyploid banana arose from naturally occurring interspecific hybrids between

banana and plantain (Perrier et al., 2011). The genomic contributions from these two species determine the ploidy and genetic make-up or subgroups. For example, a "true plantain" is a triploid banana with two *acuminata* (A) genomes and one *balbisiana* (B) genome (AAB). Other natural hybrid subgroups include "cooking banana" (ABB) and human-made or synthetic tetraploid hybrids (AABB) (Bioversity International, 2018; Irizarry, et al., 2001). Culturally, plantain has been categorized as three flowering/bunch-producing types, including French (persistent bracts and male flower bud, with many hands), false horn (degenerate male bud, intermediate number of hands), and horn (no male bud, few hands) (Bioversity International, 2018).

Although plantain is grown in most tropical regions of the world, the Democratic Republic of Congo, Cameroon, and Ghana in Central and Western Africa and Colombia in South America are the largest producers, with total productions of 4,800,000, 4,535,978, 4,050,630, and 3,575,706 t, respectively (Food and Agriculture Organization of the United Nations, 2018). In Puerto Rico, plantain is an important cash crop with an annual farm value of \$72.4 million (Puerto Rico Institute of Statistics, 2018), and it is planted on ≈23,000 acres (U.S. Department of Agriculture, 2012).

BLSD, which is commonly known as black sigatoka, is a leaf spot disease caused by the plant pathogenic fungal anamorph *Pseudocercospora fijiensis* (teleomorph *Mycosphaerella fijiensis*). Since its description in 1963, BLSD has spread to most banana and plantain growing regions. BLSD in Latin America was first recorded in Honduras in 1972; since then, the disease has spread to Belize (1975), Guatemala (1977), Nicaragua, El Salvador and Costa Rica (1979), Mexico (1980), Panama and Colombia (1981), Ecuador (1986), Venezuela (1991), Peru (1994), Bolivia (1996), Brazil (1998), Bahamas (2004), and French Guiana (2008). In the Caribbean region, the disease was first reported in Cuba (1992), Jamaica (1994), Dominican Republic (1996), Haiti (2000), Trinidad and Tobago (2003), Grenada (2005), Saint Vincent (2009), St. Lucia and Martinique (2010), and Dominica and Guadeloupe (2012)

(Robinson and Galán-Saúco, 2010). In the continental United States, BLS D was first found in Florida in 1998 (Ploetz and Mourichon, 1999), and it was confirmed in the U.S. territory of Puerto Rico in 2004 (Irish et al., 2006).

BLS D does not immediately kill its host plants; instead, it causes severe leaf necrosis that results in reduced photosynthetic area, thereby impacting the bunch weight and fruit quality. Without chemical and cultural control, yields can be reduced by 20% to 80%, depending on disease severity (Churchill, 2011; Etebu and Young-Harry, 2011). Fruits harvested from heavily infected plants ripen prematurely and unevenly, resulting in unmarketable fruit. The need to manage the disease to a level that does not affect fruit production and quality makes BLS D the most economically important leaf disease of plantain. In Puerto Rico, BLS D has steadily led to higher production costs associated with added labor for cultural practices and increased use of fungicides (Cortés et al., 2009; Irish et al., 2006). Approximately 30% of the total cost of production is associated with pruning of heavily infected, nonfunctional leaves and the use of chemical control measures for BLS D (Etebu and Young-Harry, 2011). The presence of BLS D at plantain farms in Puerto Rico has increased the number of fungicide applications that are needed to maintain profitable yields (Alamo et al., 2007).

Because managing BLS D with fungicidal applications can be expensive and hazardous to human health and the environment, breeding for disease resistance is the most sustainable and effective means of management. Several international breeding programs have released BLS D-resistant plantain and banana hybrids, most of which are parthenocarpic tetraploids generated by crossing triploid banana and fertile true plantain cultivars with wild or improved diploid (*Musa* AA) parents (Ortiz and Vuylsteke, 1998; Rowe and Rosales, 1993; Swennen and Vuylsteke, 1993; Vuylsteke and Ortiz, 1995).

The U.S. Department of Agriculture, Agricultural Research Service (ARS), Tropical Agriculture Research Station (TARS) in Mayaguez, Puerto Rico, is the official repository for *Musa* species germplasm for the National Plant Germplasm System. In this capacity, the authors have used *Musa* to introduce breeding programs involving disease-resistant plantain genotypes for field evaluation (Irish et al., 2013). In this study, the authors compared performance by measuring yield and fruit quality traits in two plantain cultivars grown under BLS D pressure at two locations in Puerto Rico.

Materials and methods

This study was conducted from 2011 to 2013 in Puerto Rico at the U.S. Department of Agriculture Agricultural Research Service Tropical Agriculture Research Station research farm in Isabela (Coto clay: clayey, kaolinitic isohyperthermic Typic Hapludox) and at the Corozal Agricultural Experiment Station of the University of Puerto Rico (Corozal clay: clayey, mixed, isohyperthermic

Aquic Haplohumults). Soil and climatic characteristics are described in Tables 1 and 2, respectively. Soil samples from each site were collected 2 months before planting by taking 15 borings at a depth of 0–25 cm from each of the projected cultivar rows. Samples were air-dried and passed through a 16-mesh screen. Soil pH in water and 0.01 M calcium chloride (1 soil: 2 water) were measured with a glass electrode. Phosphorous (P) and exchangeable cations [potassium (K), magnesium (Mg), and calcium (Ca)] were extracted with Mehlich III solution [extract was composed of 0.2 M acetic acid, 0.25 M ammonium nitrate, 0.015 M ammonium fluoride, 0.013 M nitric acid, and 0.001 M ethylene diamine tetraacetic acid (EDTA) (Amacher, 2007)] and determined by inductively coupled plasma (ICP) spectrometry (Sumner and Miller, 2007). Organic carbon was determined using the Walkley-Black method (Nelson and Sommers, 2007). Soil ammonium (NH₄) and nitrate (NO₃) were determined by steam distillation (Mulvaney, 2007).

The plantain cultivars evaluated in the experiment were the French type, which is a BLS D-resistant FHIA-21 “cooking banana” (*Musa* AAAB), and the false-horn type, which is a disease-susceptible Maricongo “true plantain” (*Musa* AAB). ‘FHIA-21’ was developed at the Fundación Hondureña de Investigación Agrícola (FHIA) in Honduras (Central America). ‘Maricongo’ is the most extensively grown cultivar that is commercially produced in Puerto Rico. The ‘FHIA-21’ source planting material was obtained as virus-free (indexed) in vitro-cultured plantlets from the Bioversity International’s

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Units			
To convert U.S. to SI, multiply by	U.S. unit	SI unit	To convert SI to U.S., multiply by
0.4047	acre(s)	ha	2.4711
100	bar	kPa	0.01
0.3048	ft	m	3.2808
2.54	inch(es)	cm	0.3937
25.4	inch(es)	mm	0.0394
0.4536	lb	kg	2.2046
1.1209	lb/acre	kg·ha ⁻¹	0.8922
28.3495	oz	g	0.0353
28,350	oz	mg	3.5274 × 10 ⁻⁵
0.001	ppm	mg·g ⁻¹	1000
1	ppm	mg·kg ⁻¹	1
0.9072	ton(s)	t	1.1023
(°F – 32) ÷ 1.8	°F	°C	(°C × 1.8) + 32

Table 1. Average preplant soil characteristics at two plantain test sites in Puerto Rico measured to a depth of 25 cm (9.8 inches).

Soil characteristics ^z	Corozal (Ultisol)	Isabela (Oxisol)
pH in water	4.63	6.36
pH in calcium chloride	4.0	5.77
Ammonium nitrogen (mg·kg ⁻¹)	386	41
Nitrate nitrogen (mg·kg ⁻¹)	28	22
Organic carbon (%)	1.28	1.52
Phosphorous (mg·kg ⁻¹)	9	30
Potassium (mg·kg ⁻¹)	133	143
Calcium (mg·kg ⁻¹)	1645	1625
Magnesium (mg·kg ⁻¹)	62.5	95
Aluminum (mg·kg ⁻¹)	200	ND ^y

^z1 mg·kg⁻¹ = 1 ppm.^yND = not detectable.**Table 2. Weather data at two plantain test sites in Puerto Rico (2011–13).**

Site characteristics ^z	Corozal (Ultisol)	Isabela (Oxisol)
Total rainfall (cm)	357	326
Maximum temperature (°C)	29.3	28.5
Minimum temperature (°C)	20.8	21.4
Elevation (m)	195	126

^z1 cm = 0.3937 inch, (1.8 × °C) + 32 = °F, 1 m = 3.2808 ft.

Musa International Transit Center at the Catholic University in Leuven, Belgium. Before field transplanting, 'FHIA-21' plant material was increased via in vitro micropropagation following conventional protocols (Vuylsteke, 1989) and acclimatized in a greenhouse for 3 months. Plants were regularly watered and fertilized with 20N–8.7P–16.6K water-soluble fertilizer (Evergreen + micro elements; Plant Foods, Vero Beach, FL) and 19N–2.6P–10K slow-release fertilizer (Osmocote; Scotts Co., Marysville, OH). Six-month-old tissue-cultured plantlets of 'FHIA-21' and medium-size corms obtained from sword suckers of 'Maricongo' were planted at 6 × 6-ft spacing in a randomized complete block design with six replications. Within a replication, there were two plots of two rows to accommodate each treatment (cultivars). Each treatment plot had 14 plants, of which the inner 10 were harvested. Alleys of 12 ft separated the replications (equivalent to 806 plants/acre). Two guard plants of each cultivar separated the 10 experimental treatment plants between treatment plots, and there were two guard rows at the end of each plot. The experiment was surrounded by three rows of BLSD-susceptible 'Grande Naine' Cavendish subgroup banana (*Musa* AAA) plants that served as a source of natural inoculum for *P. fijiensis*.

Fertilization was provided using a 10N–2.2P–24.9K–1.8 Mg granular commercial mixture by applying one-quarter pound and one-half pound at 1 and 2 months after transplanting, respectively. One pound of fertilizer per plant was used thereafter every 3 months until the experiment was completed. Nematodes, soil-borne insects, and weeds were controlled by following recommended cultural practices (Agricultural Experiment Station, 1995). Plots were drip-irrigated when the soil water tension, measured with tensiometers at a depth of 1 ft, exceeded 20 kPa. No leaf pruning or foliar spraying was performed to control BLSD. A selective desuckering program was implemented ≈4 months after planting to allow the development of a single ratoon sucker per stump for the subsequent crop.

Plantain fruits are marketed by units after considering size and weight. Larger and heavier fruit command premium prices. In Puerto Rico, a plantain fruit is graded as marketable if its weight is more than 270 g. Assuming that proper cultural practices are used, this criterion is easily achieved by fruit harvested from false-horn cultivars such as Maricongo. 'FHIA-21' is a French-type plantain producing bunches with as many as nine hands; however, some of

these hands produce small unmarketable fruit. To increase individual fruit size and weight (>270 g), the number of fruit-bearing hands was reduced for this cultivar. The practice consisted of the removal of the floral bud and lower hands of the immature bunches, maintaining only the four uppermost hands in the bunch. The false-horn bunches of 'Maricongo' were left unpruned to produce their approximate average number of seven hands. In previous experiments, the authors have demonstrated that growing French-type plantain cultivars under intensive management and pruning of the male floral bud and lower hands in the immature bunch increased individual fruit size and weight. Those results showed a 20% increase in marketable fruit when compared with the false-horn 'Maricongo' (Goenaga and Irizarry, 2006; Irizarry and Goenaga, 1995, 1997; Irizarry et al., 1998).

At flowering (bunch-shooting) and harvest, the number of functional leaves per plant was recorded. The number of days to flower was calculated as the time interval between field planting and flowering in the plant crop; in the ratoon crop, it was calculated as the number of days that elapsed between plant crop harvest date and the ratoon bunch shooting date. At flowering, plant height and pseudostem diameter of the mother and ratoon plants were measured. These measurements were taken from the base of the plants to the point of bunch emergence for the plant height, and to 1 m above ground level for the pseudostem diameter.

Bunches were harvested when the fruits reached the mature green stage, which occurred ≈110 d after flowering. At harvest, the bunches were weighed, and the numbers of hands and fruit were counted and then cut from the rachis. Fruit outer length and diameter at the widest point were measured from three fruits of the middle section of the third upper and last hands in the bunch. These fruit measurements were pooled to obtain an average for each hand. The weights of both the third hand and last hand were also recorded. Values for bunch weight and yield per area were obtained after subtracting the rachis weight from the total bunch weight.

The severity of BLSD was assessed at flowering time for the plant and

ratoon crops. Using Gauhl's modification of Stover's severity scale adapted from Orjeda (1998), all leaves for every test plant were scored on a 0–6 scale (0 = no symptoms; 1 = 1%; 2 = 2% to 5%; 3 = 6% to 15%; 4 = 16% to 33%; 5 = 34% to 50%; and 6 = >51% of the leaf area affected).

To determine the presence of *P. fijiensis* causing infection on evaluated plantain cultivars from other possible related fungal species, 20 isolates were recovered for testing. Single ascospore cultures were isolated from randomly selected senescent leaf tissue from five 'Maricongo' and five 'FHIA-21' plants from both the Corozal and Isabela locations using a discharge technique described by Johanson and Jeger (1993). Fungal mycelium was grown in potato dextrose agar broth on an orbital shaker, harvested, and lyophilized before DNA extraction. DNA was extracted using a DNeasy Plant Mini Kit (Qiagen, Valencia, CA) following the manufacturer's protocols for fungi. DNA for each isolate was amplified using a *P. fijiensis* species-specific primer set (ACTR/MfactF) in combination with a positive internal control primer set (TMG3/TMG4) for the β -tubulin gene developed by Arzanlou et al. (2007). Polymerase chain reactions (PCRs) also included genomic DNA from pure cultures of *P. fijiensis*, *P. musicola* (yellow sigatoka), and *P. eumusae* (eumusae leaf spot) (provided by G. Kema) as well as a negative water control to serve as references. All PCRs were performed as described by Arzanlou et al. (2007) on a thermocycler (T-100; Bio-Rad, Hercules, CA) and were electrophoresed in 1.5% agarose gels and stained with ethidium bromide. Agarose gels were visualized and photographed using a transilluminator (Epi Chemi II Darkroom; UVP, Upland, CA).

To determine soluble sugars and starch, 500-mg fruit samples were collected at harvest time (mature green stage) and ground after lyophilization. A fruit sample subset was allowed to fully ripen (ripened yellow stage) and analyzed for sugars and starch. Soluble sugars were extracted with deionized water at 60 to 75 °C for 1 h. Glucose, fructose, and sucrose concentrations were determined using GAHK-20 (glucose), FA-20 (fructose), and SCA-20 (sucrose) assay reagent kits. All kits were purchased

from Sigma-Aldrich (St. Louis, MO). Starch concentration was determined with a commercial enzymatic starch assay kit (SA-20; Sigma-Aldrich), and all steps were followed in accordance with the technical instructions provided by the manufacturer.

Analysis of variance was performed using the GLM procedure of SAS (version 9.4 for Windows; SAS Institute, Cary, NC). After a significant F test at $P \leq 0.05$, mean separation was performed with Tukey's honestly significant difference range test.

Results and discussion

To confirm the presence of *P. fijiensis* on the evaluated plantain cultivars, 20 isolates were recovered for testing. Using an ascospore discharge technique (Johanson and Jeger, 1993), single ascospore cultures were recovered from randomly selected senescent leaf tissue from five 'Maricongo' and five 'FHIA-21' plants from both the Corozal and Isabela locations. Isolates were tested with a *P. fijiensis* species-specific primer set in combination with a positive internal control β -tubulin gene primer set. Following PCR amplification, *P. fijiensis* infection was confirmed at both locations by electrophoretic separation and visualization of both the diagnostic (≈ 500 bp) and β -tubulin gene (≈ 300 bp) fragments from isolates recovered from both cultivars as well as from the positive control reference sample. All 10 isolates recovered from both cultivars from Isabela amplified the diagnostic ≈ 500 -bp and internal control fragments. In contrast, only two isolates/DNA samples from 'FHIA-21' and one from 'Maricongo' amplified the ≈ 500 -bp and internal control fragments from Corozal. The remaining seven samples from Corozal amplified the internal β -tubulin gene fragment, but nothing else. In addition, only the internal β -tubulin gene fragment was amplified for both *P. musicola* and *P. eumusae* reference samples; no amplification was seen for the negative water control. These findings provided evidence of the presence of BLSD at both locations.

Year, location, and cultivars showed highly significant effects ($P \leq 0.01$) for most parameters measured in the study (Tables 3 and 4). The location $\times$ cultivar interaction was mostly not significant, indicating

that cultivars responded similarly for yield, fruit traits, plant attributes, and reaction to BLSD at both locations. A similar response was observed for the year $\times$ location, location $\times$ cultivars, and year $\times$ cultivar interactions, particularly for yield parameters.

Overall, cultivars did not exhibit drastic yield differences between the plant (year 1) and ratoon (year 2) crops (Tables 3 and 5). A common limitation of perennial plantain production is the rapid yield decline that occurs after harvest of the plant crop (Goenaga, et al., 1993; Vuylsteke et al., 1993). This yield decline "syndrome" only became apparent in this experiment after harvest of the first ratoon crop. Thereafter, the second ratoon (year 3) suckers developed a poor root system that caused plant lodging early in the growth cycle (data not shown). An exception to this problem was the performance of 'Maricongo' at Isabela, where a second ratoon crop was partially obtained. Average bunch weight and fruit per bunch for the second ratoon crop at this location were 16.4 kg/plant and 52.4 fruit/plant, respectively (data not shown). Although these yield figures are acceptable, particularly for a second plantain ratoon crop, only 47% of the experimental plants did not exhibit plantain decline symptoms and were able to be harvested. In other words, based on area, yield declined by more than 50%. The causes of this yield decline in plantain production are not well-understood, and the decline is contrary to what is observed in banana production, for which ratoon crops have higher yields than the plant crop and remain productive for several years (Goenaga and Irizarry, 1998, 2000; Vuylsteke et al., 1993). Although the plantain decline syndrome may have limited production of a second ratoon crop for both cultivars, expression of BSV symptoms were present in plants of cultivar FHIA-21, thereby possibly serving as a compounding effect for the yield decline. BSV is transmitted by mealybugs or through stress activation processes of viral sequences known to be integrated into the genome of 'FHIA-21' (Irish et al., 2013; Irizarry et al., 2001; Krikorian et al., 1998; Teycheney et al., 2005). These results confirmed that production of multiple ratoon crops is a limitation to plantain production and deserves more research attention.

Table 3. Yield and bunch traits of ‘FHIA-21’ and ‘Maricongo’ plantain planted at two locations in Puerto Rico under black leaf streak disease pressure. Values are means of six replications and 2 years (2011–13).

Location	Cultivar	Bunch yield (kg·ha ⁻¹) ^z	Fruit (no./ha) ^z	Third hand wt (kg) ^y	Third hand fruit length (cm) ^y	Third hand fruit width (mm) ^y	Last hand wt (kg) ^y	Last hand fruit length (cm) ^y	Last hand fruit width (mm) ^y	Mean fruit wt (g) ^z
Corozal	FHIA-21	26,112	99,948	3.06	24.3	40.3	2.72	22.9	39.3	258.1
	Maricongo	20,049	76,427	1.75	25.0	42.6	0.72	21.6	39.9	263.0
	Average	23,081	88,187	2.40	24.6	41.5	1.72	22.2	39.6	260.5
	HSD	4,367	6,603	0.47	NS	1.6	0.39	1.06	NS	NS
	(0.05) ^x									
Isabela	FHIA-21	41,228	122,522	4.78	27.0	43.7	4.01	25.5	42.2	335.8
	Maricongo	30,863	95,297	2.52	27.2	44.8	0.92	22.7	41.5	323.6
	Average	36,046	108,910	3.65	27.1	44.2	2.47	24.1	41.9	329.7
	HSD	1,778	4,364	0.19	NS	0.92	0.20	0.6	0.45	8.4
	(0.05)									
	HSD	2,207	3,705	0.24	0.61	0.88	0.20	0.57	0.78	15.6
	(0.05) ^w									
	Year (Y)	*	NS	*	NS	NS	NS	NS	NS	NS
	Location	***	***	***	***	***	***	***	***	***
	(L)									
	Y × L	*	NS	NS	NS	NS	NS	*	*	*
	Cultivar	***	***	***	NS	***	***	***	NS	NS
	(C)									
	L × C	NS	NS	***	NS	NS	***	*	NS	NS
	Y × C	NS	NS	*	*	**	NS	**	***	**
	Y × L × C	NS	NS	NS	NS	NS	NS	NS	NS	NS

^z1 kg·ha⁻¹ = 0.8922 lb/acre, 1 fruit/ha = 0.4047 fruit/acre, 1 g = 0.0353 oz.

^yThe first three upper hands of plantain bunches contain ≈50% of the total fruits. Therefore, the weight of the third upper hand is often used to represent an average weight of all the hands in the bunch. Similarly, higher fruit weight and size in the last hand are indicative of higher marketable weight of fruits of hands after the third hand (Goenaga and Irizarry, 2006); 1 kg = 2.2046 lb, 1 cm = 0.3937 inch, 1 mm = 0.0394 inch.

^xTukey's honestly significant difference test at $P = 0.05$.

^wCompared means among locations.

NS, *, **, ***Nonsignificant or significant at $P \leq 0.05$, 0.01, or 0.001, respectively.

Total number of fruit, as well as many other bunch and hand traits were significantly different between locations (Table 3). The Corozal location is considered part of the plantain-growing region in Puerto Rico; however, significantly less fruit were produced at Corozal (88,187 fruit/ha) than at Isabela (108,910 fruit/ha) (Table 3) over two cropping cycles. Less productivity at Corozal than at Isabela could have been due to a combination of factors. First, BLSD severity was significantly higher at Corozal than at Isabela (Table 6). There were numerous unmanaged banana and plantain plantations near the Corozal experimental site and as a result, perhaps inoculum levels and disease pressure were higher at this location than at Isabela. Averaged over all leaves across both cultivars, BLSD severity was 2.76 at Corozal and 1.61 at Isabela (Table 6), thus explaining the significant location effect. However, a reading of 2.76 meant that only ≈2% to 5% of the leaf

area was affected by the disease. Values close to 4.0 (16% to 33% of leaf area affected) were observed in older leaves (leaves 7–12) at Corozal (Table 6). However, even this severity in older leaves may not fully explain significantly less productivity at Corozal than at Isabela (Table 3). Studies with banana (Robinson et al., 1992) and plantain (Cayón, et al., 1995) have shown higher rates of photosynthesis in younger leaves (leaves 2–7) and concluded that older leaves contributed very little to fruit filling. Hence, a second reason for lower productivity at Corozal may be related to soil factors interfering with optimum nutrient uptake. The soil at Corozal is highly weathered Ultisol, typical of the humid tropics. These soils used for growing plantain are characterized by intensive leaching, with moderate to low cation exchange capacity, high soil acidity, and, consequently, a low supply of bases (U.S. Department of Agriculture, 1999). The soil at the Corozal

site is more acidic than that at Isabela (Table 1). Magnesium deficiency is a constraint to crop productivity in acidic soils due to the high saturation of the soil cation exchange capacity with hydrogen (H⁺), thus causing Mg leaching and impaired plant uptake (Gransee and Führes, 2013). Because highly weathered soils are usually intensively limed, “Mg fixation,” which is caused by the co-precipitation of Mg with aluminum (Al) upon liming, may have a major role in limiting the availability of Mg in crops grown on these soils (Irizarry et al., 2000; Martínez et al., 1996). Mg is required for chlorophyll synthesis, and critical enzymes and enzymatic reactions require this element for optimum metabolism. In addition, Mg is involved in carbohydrate transport from source-to-sink organs (Farhat et al., 2016). Therefore, optimal growth and productivity are impaired in Mg-deficient plants. Leaf tissue sampling conducted during the experimental period showed an average Mg

Table 4. Plant traits of ‘FHIA-21’ and ‘Maricongo’ plantain planted at two locations in Puerto Rico under black leaf streak disease pressure. Values are means of six replications and 2 years (2011–13).

Location	Cultivar	Plant ht (m) ^z	Stem diam (cm) ^z	Leaves at flowering (no.)	Leaves at harvest (no.)	Time to flower (d)	Time to harvest (d)
Corozal	FHIA-21	2.97	16.7	11.2	4.1	563	666
	Maricongo	3.29	16.3	9.2	0.9	518	605
	Average	3.13	16.5	10.2	2.6	540	636
	HSD	0.15	NS	0.89	0.85	38.9	44.5
	(0.05) ^y						
Isabela	FHIA-21	3.54	20.7	11.7	6.1	430	526
	Maricongo	3.7	19.2	10.1	1.8	395	487
	Average	3.6	19.9	10.9	3.9	412	506
	HSD	0.07	0.47	0.47	0.49	8.7	10.0
	(0.05) ^y						
	HSD	0.08	0.57	0.47	0.46	18.7	21.3
	(0.05) ^x						
	Year (Y)	***	***	***	NS	***	***
	Location	***	***	*	***	***	***
	(L)						
	Y × L	NS	NS	NS	***	NS	NS
	Cultivar (C)	***	**	***	***	***	***
	L × C	*	NS	NS	*	NS	NS
	Y × C	***	NS	NS	***	NS	NS
	Y × L × C	**	*	NS	NS	NS	NS

^z1 m = 3.2808 ft, 1 cm = 0.3937 inch.

^yTukey's honestly significant difference test at $P = 0.05$.

^xCompared means among locations.

NS, *, **, ***Nonsignificant or significant at $P \leq 0.05$, 0.01, or 0.001, respectively.

Table 5. Bunch yield and number of fruit of ‘FHIA-21’ and ‘Maricongo’ plantains grown at two locations in Puerto Rico under black leaf streak disease pressure. Values are means of six replications.

Cultivar	Corozal			Isabela		
	2-yr	Plant crop	Ratoon crop	2-yr	Plant crop	Ratoon crop
	Bunch yield (kg·ha⁻¹)^z					
FHIA-21	26,112	26,611	25,614	41,228	44,850	37,607
Maricongo	20,049	17,740	22,358	30,863	32,989	28,736
HSD ^y	4,367	3,938	NS	1,778	3,771	1,614
	Fruit yield (no./ha)^z					
FHIA-21	99,948	99,632	100,264	122,522	123,918	121,127
Maricongo	76,427	77,341	75,513	95,297	98,602	91,991
HSD	6,603	7,455	13,288	4,364	7,724	6,460

^z1 kg·ha⁻¹ = 0.8922 lb/acre, 1 fruit/ha = 0.4047 fruit/acre.

^yTukey's honestly significant difference test at $P = 0.05$.

leaf concentration of 0.25% ($n = 72$ samples) for cultivars grown at Corozal (data not shown), whereas the average leaf Mg concentration at Isabela was 0.34% ($n = 34$). Studies conducted by Irizarry et al. (1988, 1990, 2000) showed that Mg concentration of 0.30% in leaves of banana and plantain grown in Ultisol is considered optimum for growth and productivity. Hence, limited Mg uptake may further explain reduced productivity at Corozal. It may also help to explain shorter plants with

smaller stem diameters and more days required to flower at the Corozal site (Table 4). Because of the high-K fertilization requirement for plantain, and because K has an antagonistic effect with Mg, it is important for growers to monitor leaf Mg concentrations throughout the growing season, particularly if the crop is established in a weathered soil typical of the humid tropics. Furthermore, nonproductive plants in old plantations may serve as a source of disease inoculum;

therefore, they should be eliminated before the establishment of a new field nearby.

At both locations, ‘FHIA-21’ produced significantly more fruit and yield per area than ‘Maricongo’ (Tables 3 and 6). The weights of the third-upper and last hand in the bunch were also significantly higher in cultivar FHIA-21 than in Maricongo (Table 3). Fruit length of the third-upper hand was not different between cultivars at either location, but the Maricongo fruit in this hand were significantly thicker (Table 3). In contrast, fruit in the last hand of ‘FHIA-21’ tended to be longer and thicker at both locations. Average mean fruit weight was significantly higher in at Isabela than at Corozal (Table 3). At Isabela, the mean fruit weight was significantly higher in ‘FHIA-21’ than in ‘Maricongo’, but both cultivars exceeded the local market minimum weight of 270 g to be considered marketable fruit (Goenaga and Irizarry, 2006; Irizarry and Goenaga, 1995). This is in contrast to fruit produced by both cultivars at Corozal, where the average mean fruit weight was 21% lower than that at Isabela (Table 3). The higher mean

Table 6. Black sigatoka disease severity for 'FHIA-21' and 'Maricongo' plantain planted at two locations in Puerto Rico. Values are means of six replications and 2 years (2011–13).

Location	Cultivar	Leaf disease severity (youngest/top most to oldest)											
		1 ^z	2	3	4	5	6	7	8	9	10	11	12
Corozal	FHIA-21	0	0.10	0.61	1.34	1.95	2.62	3.00	3.34	3.89	4.07	3.94	4.41
	Maricongo	0	0.06	0.79	1.47	2.24	2.84	3.48	4.34	4.79	5.23	5.71	6.00
	Average	0	0.08	0.70	1.40	2.09	2.73	3.24	3.84	4.34	4.65	4.82	5.20
	HSD (0.05) ^y	0	NS	NS	NS	NS	NS	NS	0.61	NS	0.88	1.60	0.66
Isabela	FHIA-21	0	0	0	0.05	0.20	0.52	1.02	1.45	1.97	2.37	2.82	3.29
	Maricongo	0	0	0.02	0.32	0.72	1.47	2.27	2.92	3.52	4.10	4.70	4.87
	Average	0	0	0.012	0.18	0.46	0.99	1.64	2.18	2.74	3.23	3.76	4.08
	HSD (0.05)	0	0	NS	0.17	0.20	0.30	0.32	0.39	0.35	0.34	0.50	0.50
	HSD (0.05) ^x	0	0.07	0.16	0.17	0.23	0.39	0.35	0.33	0.48	0.39	0.49	0.25
	Year (Y)	NS	NS	NS	**	**	**	**	**	*	**	**	NS
	Location (L)	NS	*	***	***	***	***	***	***	***	***	***	***
	Y × L	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
	Cultivar (C)	NS	NS	NS	*	**	**	***	***	***	***	***	***
	L × C	NS	NS	NS	NS	NS	NS	*	NS	NS	NS	NS	*
	Y × C	NS	*	NS	NS	NS	NS	NS	NS	NS	NS	NS	**
	Y × L × C	NS	*	NS	NS	NS	NS	NS	NS	NS	**	NS	*

^zDisease severity based on a 0–6 rating scale (0 = no symptoms; 1 = 1%; 2 = 2% to 5%; 3 = 6% to 15%; 4 = 16% to 33%; 5 = 34% to 50%; and 6 = >51% of the leaf area affected) found in Orjeda (1998).

^yTukey's honestly significant difference test at $P = 0.05$.

^xCompares means among locations.

NS, *, **, ***Nonsignificant or significant at $P \leq 0.05$, 0.01, or 0.001, respectively.

Table 7. Concentrations of starch and soluble sugars at harvest time (mature green) and fully mature (ripened yellow) fruit of 'FHIA-21' and 'Maricongo' plantain. Values are means of four replications of fruit grown at Isabela, PR.

Cultivar	Harvest time				Fully mature fruit			
	Starch	Glucose	Fructose	Sucrose	Starch	Glucose	Fructose	Sucrose
	(mg·g ⁻¹) ^z				(mg·g ⁻¹)			
FHIA-21	378.2	ND ^y	0.925	6.0	26.1	72.2	43.9	185.5
Maricongo	522.5	ND	2.00	4.1	164.5	54.2	68.9	320.2
HSD (0.05) ^x	NS	NS	NS	NS	40.7	8.7	3.4	41.1

^z1 mg·g⁻¹ = 1000 ppm.

^yND = not detectable.

^xTukey's honestly significant difference test at $P = 0.05$; NS = nonsignificant.

fruit weight obtained at Isabela for 'FHIA-21' was 23% higher than that obtained for French-type 'Superplatan' planted in the fertile, semi-arid region of Puerto Rico under drip irrigation (Irizarry and Goenaga, 1995) or 24% higher than the mean fruit weight of 'Maiden', which is a promising French-type cultivar evaluated at Isabela (Goenaga and Irizarry, 2006).

The number of functional leaves present at flowering is an important physiological trait for proper fruit-filling because no new leaves are produced after this stage. It is generally accepted that eight leaves at flowering time are sufficient for adequate bunch growth and development (Robinson et al., 1992; Vargas, et al., 2009). If the incidence of BLSD is high at this stage, then bunch-filling will be curtailed and yield will be reduced

dramatically as a result of reduced photosynthetic area. At both locations, the number of leaves at flowering was significantly higher in 'FHIA-21' than in 'Maricongo' (Table 4). Furthermore, at harvest time, 'FHIA-21' plants had significantly more functional leaves than 'Maricongo'. This indicated that during the ≈3-month period between flowering and harvest, 'FHIA-21' plants had more leaf photosynthetic area for adequate bunch-filling, possibly contributing to the higher yield of this cultivar at both locations. It also confirmed good BLSD resistance in 'FHIA-21'.

Plantain is usually consumed unripe (starchy) and is prepared by frying, boiling, or steaming; however, it can be consumed ripe (sweet) and prepared by frying or roasting. When harvested green (unripe), the concentration

of starch is high; as the fruit matures, starch is converted to sugars and the peel color changes from green to yellow. Little is known about the organoleptic characteristics of 'FHIA-21'. To be accepted by consumers, organoleptic traits of 'FHIA-21' should be like those of commercial cultivars, particularly the false-horn standard commercial cultivar Maricongo. In this study, no significant differences were found for starch and soluble sugars in green unripe fruit between cultivars (Table 7). For fully mature fruit, the concentration of starch in 'FHIA-21' was significantly lower than that in 'Maricongo', and most soluble sugars were significantly higher. This trait of 'FHIA-21' may be desirable because many consumers prefer sweeter plantain.

In conclusion, this study showed that 'FHIA-21' has good resistance

against BLSD and is a viable alternative to current disease-susceptible cultivars. At the current (2019) farm gate price of \$0.35 per fruit, the average fruit yield of 'FHIA-21' (Table 5) would have amounted to gross profits of \$34,982 and \$42,883 per hectare at Corozal and Isabela, respectively, without the need to incur fungicide application costs. The threat of 'FHIA-21' being affected by BSV, particularly during periods of stress (Dallot, et al., 2001; Hauser, 2010), should not be discounted, and efforts should be made to provide optimum agronomic management to prevent potential stressors.

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