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## Tackling black leaf streak disease and soil fertility constraints to enable the expansion of plantain production to grassland in the humid tropics

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In Central Africa, plantain is traditionally grown after a forest fallow. Given increasing urban demand and a lack of forest fallows near urban centres, as well as poor roads and environmental concerns to reduce pressure on forests, research is needed to identify suitable shade, fertility management and cultivars to shift production of plantain to grasslands and to reduce losses to diseases such as black leaf streak disease (BLS). Effects of light level (full, 67%, 33% light), and nitrogen (N)-amendment on BLS-tolerant (FHIA-21) and BLS-susceptible (Batard) cultivars planted on soil from paired grassland and forest sites were determined. BLS and growth were monitored until 5 months after planting. Three months after planting, leaf area attacked on cultivar FHIA-21 was less than half that on Batard. Plants grown under 33% and 67% light had less leaf area attacked (2.9% and 4.6%, respectively) than those grown in full light (7.3%). Leaf area and dry matter (DM) were higher under shade and when grown on forest soils. Compared to growing BLS-susceptible plantain on forested land under shade, a shift onto grasslands and a reduction in shade use is predicted to reduce yields. Using cultivar FHIA-21 may limit, but not eliminate, yield loss.

**Keywords:** Black leaf streak disease; black sigatoka; FHIA-21; *Musa* spp. AAB; *Mycosphaerella fijiensis*; plantain; shade

### 1. Introduction

In southern Cameroon, plantain (*Musa* spp. cv. AAB) is the most important food cash crop and is a favoured staple. Traditionally, plantain has been grown after a secondary forest fallow, with shade-trees retained, in intercropping systems. Chemical inputs (fertiliser, herbicides, pesticides) are rarely used, and burning is the most common method of field preparation. In general, only the planted crops are harvested, as fields are abandoned before the ratoons produce bunches. Recently, plantain production has become more market-orientated and seasonal price fluctuations reflect an insufficient supply to urban markets (Nkenda and Akyeampong 2003). In 2009, the total production of bananas in Cameroon was estimated to be 1 million megagrams (Mg), while total plantain production was 1.6 million Mg (FAO 2009). For Africa, the annual production of bananas and plantains exceeds 37 million Mg (FAO 2009).

Given greater urban demand, farmers aim to grow plantain nearer to the cities: (i) where forested land is rare but grassland and short-fallow land are readily available (Hauser et al. 2008), and (ii) in a more intensive manner, given easier access to fertiliser stores than in rural areas.

Recent work (Hauser et al. 2008) demonstrated that yields of two cultivars of plantain on short fallow land were only 34% and 25% of the respective adjacent forest controls. Plantains grown on short fallow land were more responsive to fertiliser application, yet this could not balance the yield loss. Clearly, there are factors other than soil chemical fertility determining yield, one of which may have been differential effects of black leaf streak disease (BLS), also known as black sigatoka, caused by the fungus *Mycosphaerella fijiensis* (Morelet), which was not monitored in that trial (Hauser et al. 2008).

BLS is the greatest threat to the banana industry, affecting both bananas and plantains (De Lapeyre de Bellaire et al. 2010). In plantain, it can cause yield losses of 33% (Mobambo et al. 1993) as the fingers of the bunch fail to fill. In the tropics, in large plantations, successful control is achieved both by aerial fungicide application 10–60 times per year, depending on climatic conditions (Abadie et al. 2009) and by leaf-pruning. Globally, the cost of controlling BLS in export banana plantations is estimated to be US\$1000 per hectare (Arias et al. 2003).

Intensive use of fungicides is undesirable due to high cost, environmental impact and potential for the

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development of resistance in *M. fijiensis* (Romero and Sutton 1997). Because fungicide is not generally available for smallholder plantain farmers, some other means of control is required for managing the spread and impact of BLS. The development of resistant cultivars is an important component in BLS management, preferably as part of an integrated pest management programme. The resistant cultivar FHIA-21 has been developed at the Fundación Hondureña de Investigación Agrícola (FHIA), Honduras. Dzomeku et al. (2010) compared FHIA-21 to two IITA (International Institute of Tropical Agriculture) plantain hybrids, PITA-1 and PITA-4, and to local Ghanaian land races. They found that FHIA-21 had a significantly higher bunch mass and a shorter planting-to-harvest time than the other cultivars. Although there is anecdotal evidence from agroforestry trials that *Musa* grown under shade trees are less severely damaged by BLS, the link with light intensity is not well understood (Donzelli and Churchill 2007), so further research is required (Churchill 2010). The link between soil fertility and BLS incidence and severity is also unclear (van Asten et al. 2005), although it would be expected that on higher fertility soils, leaf production rates would be higher and therefore exposure to BLS would be shorter. Indeed, Okumu et al. (2011) showed that leaf production rates as well as soil K, Mg, pH and N concentrations were also positively correlated with yield.

Research is needed to assess how plantain can be grown successfully in short fallows, and to identify the requirements for optimum shade and fertility management. Here, the effects of light level (full, 67%, 33% light), and nitrogen (N)-amendment were determined on the early growth of two cultivars of plantain which were planted on soils from paired grassland and forest sites.

We used the approach of excavating soil from four different areas and transporting it to one location so we could reduce climatic and microclimatic variability and thus test the response of cultivars to soil and controlled shade only.

## 2. Material and methods

Topsoil (0–15 cm depth) was excavated from four villages in central Cameroon where the soils are classified as Ultisol, Rhodic Kandiudult (Champetier de Ribes and Aubague 1956 quoted in Koutika et al. 2008): Essong (N 04°05', E 11°35') and the contiguous Essong Mitsang (N 04°05', E 11°35'), Oboa (N 04°6', E 11°38'), and Ngoungoumou (N 04°05', E 11°40') from grassland and adjacent forest. Forests were at least 10 ha in size and at least 25 years old. In Oboa, the grassland was savannah of a mixed grass/sedge community dominated by *Imperata cylindrica* (L.) Raeusch., with a sparse shrub layer. In Ngoungoumou, *Pteridium aquilinum* L. Kuhn was also very frequent. In Essong Mitsang, the areas were *I. cylindrica*-dominated

short fallow, where *I. cylindrica* comprised more than 90% of the above-ground plant biomass.

Soil from all sites was transported in clean, 80-kg flour bags to the IITA research farm in Mbalmayo, southern Cameroon (3°51' N and 11° 27' E, 540 m a.s.l.) where mean annual rainfall is 1513 mm showing a bimodal temporal distribution. Rains start in March and end in early July, followed by a short dry season of six to eight weeks, then restart in September and stop at the end of November. Rainfall in the current year, measured as a daily average of three rainfall gauges at the IITA research farm, was 1350 mm with the months of January, July, November and December each having less than 25 mm (S. Hauser, pers. comm.). Mean annual insolation is 1645 hours. There were ongoing plantain and banana trials within 100 m of where the experiment was conducted; they were a potential source of inoculum.

Large roots were removed from the soil, then subsamples of 1 kg were taken for dry matter determination and chemical analysis. Soil (15 kg equivalent dry mass) was added to 30 L capacity circular basins of internal diameter 0.39 m and surface area 0.12 m<sup>2</sup>. Water was added so that all soils had the equivalent soil-water content. The basins were left out in the open but were buried to soil level in sawdust beds to avoid heating of their sides.

The experiment was a three-factorial, randomised complete block design, where factor one was light level at three levels; full light (100%), 67% light and 33% light established with different shade cloths. Factor two was a combined fertiliser/land-use system treatment at three levels: forest soil, grassland soil, and grassland soil amended with N fertilisation, which consisted of 17.9 g of urea per plant or 29 kg urea per hectare, assuming an average plant density of 1600 ha<sup>-1</sup>. Factor three was plantain cultivar; one of which was tolerant of BLS (FHIA-21, a “French” type hybrid) and one which was not (Batard, a Cameroonian land race of the “false horn” type).

Plantains are classified by their inflorescence and the form of the bunch. “French” plantains have a complete inflorescence, a persistent male bud, and hermaphrodite flowers. They produce bunches with many hands and many small fruits per hand. “False horn” plantains have an incomplete inflorescence, the male bud is not persistent and they have few hermaphrodite flowers. They produce bunches with fewer hands than French plantains, with fewer fruits per hand but larger fruits (Tezenas du Montcel et al. 1983). We selected FHIA-21 and Batard as they have similar average flowering heights of around 4 m. We focused on early growth, given that this correlates well with final bunch yield, as those plants that are tallest tend to produce leaves faster, flower earliest and produce the largest bunches (Swennen and de Langhe 1985). The four sources of the soils (villages) were the replicates and this was used as the blocking factor.

One plantain sucker was planted in each basin on 18 June 2005. These plantain suckers were Corm Fragment Shoots (CFS) of similar size, supplied from VESMA, Cameroon. CFS propagation refers to growing plantlets from cut corm parts, in a nursery (Lefranc et al. 2010).

Leaf production, height and circumference, together with the number of standing leaves, were monitored every 2 weeks. The position of the youngest leaf with symptoms (YLWS) was registered (after Mobambo et al. 1993). A visual BLSA assessment was done at 3 months after planting (MAP), using a scale based on Stover and Dickson (1970), modified by Gauhl (1989) and Pasberg-Gauhl (1989) and detailed in Mobambo et al. (1993) with the following classes for % leaf area ( $y$ ): 0 = without symptoms; 1 = 1% and/or up to 10 spots with a dry centre; 2 = 1–5%; 3 = 6–15%; 4 = 16–33%; 5 = 34–50%; 6 = 51–100%. The total living leaf area attacked for a plant with  $i$  living leaves was estimated using the following formula:

total leaf area

$$= \left( \sum_{\max} y \text{ for leaf 1 to leaf } i \text{ attacked (\%)} \right) / i. \quad (1)$$

Plants were harvested at 5 MAP and the leaf lamina was divided into “healthy area” and “necrotic area”, and both categories assessed using a LIC-OR LAI-2000 leaf area meter. Pathological and physiological causes of leaf damage were not distinguished. Samples were oven-dried to constant mass and dry mass was recorded. Specific leaf area (SLA) was calculated by dividing leaf area by dry leaf mass. Data were analysed in PASW (Predictive Analytics SoftWare) v18 (Norusis 2010) using a blocked, three factorial generalised linear model (GLM). Different dates were analysed separately. Significant differences are reported when

$P < 0.05$  for the main factors. Data on the percentage of leaf area attacked by BLSA were converted to a proportion then transformed such that  $y = \arcsin(\sqrt{y})$ , where  $0 < y < 1$ . Leaf count data were square root-transformed prior to analysis. Two plants that died during the experiment were excluded from the analysis as the cause of death could not be discerned. These values were treated as missing data. Regressions between plantain growth parameters and soil properties were also computed in PASW v18.

Soils were dried to constant mass then ground to 0.5 mm. The pH was determined in a water suspension at a 2 : 5 soil–water ratio. Available P was extracted by the Mehlich-3 procedure (Mehlich 1984) and determined by the malachite green colorimetric procedure (Motomizu et al. 1983). Organic C was determined by Heanes’ improved chromic acid digestion and spectrophotometric procedure (Heanes 1984). Total N was determined using the Kjeldahl method for digestion and ammonium electrode determination (Bremner and Tabatabai 1972).

### 3. Results

There were no treatment effects on average leaf emergence rates over the 5 months. However, the number of standing leaves was initially higher for the Batard cultivar (Figure 1); however, by approximately 3 MAP at the end of September, the FHIA-21 plants had more ( $P = 0.037$ ) standing leaves. The average youngest leaf with symptoms (YLWS) at 3 MAP was older ( $P = 0.022$ ) on the FHIA-21 plants (4.4) than on the Batard plants (3.7). At 3 MAP, the average area attacked by BLSA was significantly affected ( $P = 0.039$ ) by cultivar, with a lower percentage damage on FHIA-21 plants (2.9%) than on Batard plants (7.2%). Plants grown under 33%, 67% and full light had 2.9%, 4.6% and 7.3% of leaf area attacked,

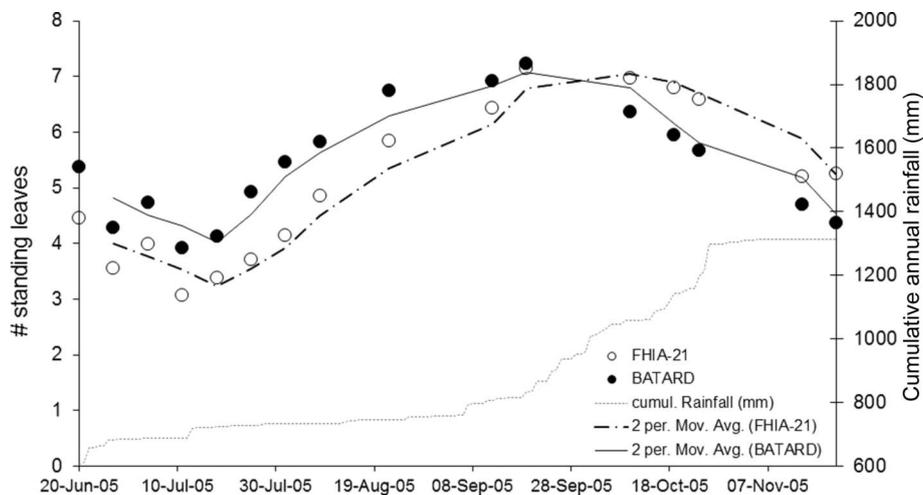


Figure 1. Pattern of number of standing leaves until 5 months after planting, separated by cultivar, and cumulative rainfall in 2005. Lines denote a moving average.

respectively, however the light factor was not quite significant ( $P = 0.068$ ).

At 5 MAP, FHIA-21 had a lower percentage ( $P = 0.022$ ) of leaf damage than Batard with 4.2% versus 8.5%, respectively. No FHIA-21 plant had more than 4% leaf damage, indeed, 50% of FHIA-21 plants had no leaf damage. All Batard plants bore some leaf damage. Damage was affected by light ( $P = 0.024$ ), with greater damage in the full light treatment than in either of the shaded treatments. However, there was no significant interaction between light level and cultivar (Figure 2).

Cultivars did not show any significant difference in growth response at 5 MAP. Specific leaf area was not different between FHIA-21 and Batard cultivars. Total leaf area and total dry matter content, averaged across cultivars, were significantly affected by light level ( $P = 0.039$  and  $P = 0.030$ , respectively) (Figure 3).

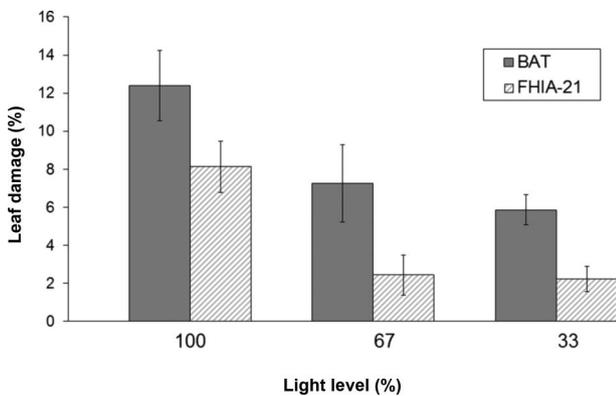


Figure 2. Percentage (%) leaf damage of plantains at 5 months after planting as affected by cultivar and light. BAT, Batard. Mean  $\pm$  SE.

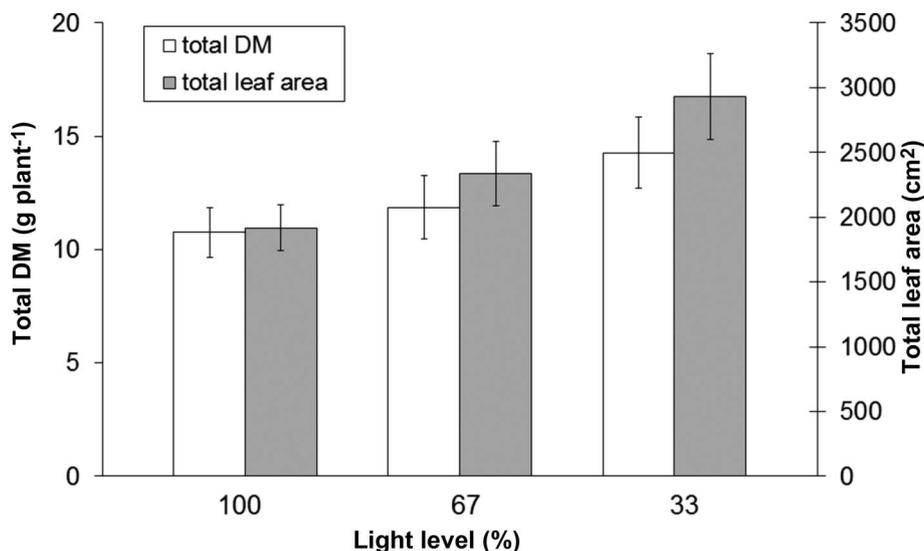


Figure 3. Total leaf area ( $\text{cm}^2 \text{ plant}^{-1}$ ) and total dry matter ( $\text{g DM plant}^{-1}$ ) at 5 months after planting as affected by light. Mean  $\pm$  SE.

Growth by 5 MAP was significantly different between forest and grassland soils (Figure 4) with plantains grown on forest soils having a greater circumference ( $P = 0.0001$ ), total dry matter ( $P = 0.0005$ ) and total leaf area ( $P = 0.0003$ ) than those on grassland soil. When fertiliser was applied to grassland, total leaf area increased to levels seen in the forest, although this was not significant; dry matter was less strongly affected ( $< 10\%$  increase) by fertiliser application.

Topsoil (0–15 cm depth) chemical parameters of grassland and forest systems in central Cameroon are given in Table 1, which shows that there were no significant differences between the land-use systems. Linear regressions between soil chemical parameters and plantain growth were not significant.

#### 4. Discussion

While the difference in early growth between FHIA-21 and Batard was not significant, this was during a time when BLSD pressure would have been low as it was the short dry season and the disease would not be expected to limit plant growth at that stage. Gauhl and Pasberg-Gauhl (1994), working in South East Nigeria, demonstrated that disease spread is via ascospores, as conidiospore counts were very low, and that ascospore production is reduced in the dry season. In the current study, once the main rains started at 3 MAP, the proportion of leaf area affected by BLSD was lower for FHIA-21 and, by 5 MAP, there was less BLSD leaf damage and thus more healthy leaf area on FHIA-21 plants. Others studies have pointed out that successful infection was promoted by extended periods of high humidity and the presence of free water on leaves (Mayorga 1990). It is therefore likely that such

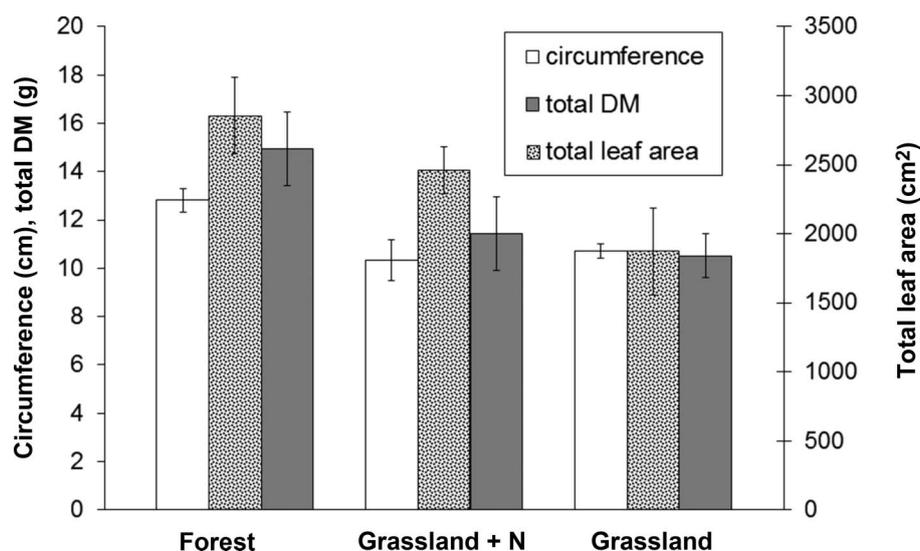


Figure 4. Effects of soil origin and urea application on plantain circumference, total dry matter (DM) and total leaf area at 5 months after planting. Mean  $\pm$  SE.

Table 1. Chemical concentrations in topsoil (0–15 cm depth) in two land-use systems (forest and grassland) in four villages in central Cameroon. No differences between land-use systems were significant.

Village	Land-use system	pH (water)	Ca <sup>2+</sup>	Mg <sup>2+</sup>	Al	K <sup>+</sup>	Mehlich available P	C org	Total N
			cmol(+) kg <sup>-1</sup>				(ppm)	(%)	(%)
Essong	Grass	6.28	4.35	1.94	0.004	0.192	7.54	1.96	0.151
Essong	Forest	5.73	4.56	2.61	0.004	0.228	7.03	2.45	0.229
E. M.	Grass	5.67	6.89	2.83	0.279	0.158	18.39	2.71	0.234
E. M.	Forest	5.35	4.61	1.89	0.059	0.099	6.86	2.61	0.188
Oboa	Grass	5.25	1.72	0.42	0.004	0.112	7.39	2.45	0.170
Oboa	Forest	4.95	1.42	0.63	0.022	0.117	3.37	2.12	0.172
Ngoungoumou	Grass	5.32	2.43	1.15	0.003	0.157	14.53	2.17	0.127
Ngoungoumou	Forest	5.83	5.21	1.42	0.004	0.114	8.09	1.82	0.144
<b>Mean</b>	<b>Grass</b>	<b>5.63</b>	<b>3.85</b>	<b>1.58</b>	<b>0.0725</b>	<b>0.155</b>	<b>11.96</b>	<b>2.32</b>	<b>0.170</b>
<b>Mean</b>	<b>Forest</b>	<b>5.47</b>	<b>3.95</b>	<b>1.64</b>	<b>0.022</b>	<b>0.139</b>	<b>6.34</b>	<b>2.25</b>	<b>0.183</b>

E. M. = Essong Mitsang village.

differences between cultivars would be cumulative and, at a later stage, would be more pronounced. The greater amount of leaf senescence at 5 MAP in the Batard cultivar supports this hypothesis, however, this outcome could also be due to differing physiological factors, which were not measured in this trial. Given that BLSD is associated with high yield losses (Mobambo et al. 1993), FHIA-21 would be expected to have produced heavier bunches.

The effects of shade on growth variables could be due to both the direct effect of reduced light on the plantain plant and an indirect effect on BLSD. Norgrove (1998), reviewing the literature on the effects of shade on *Musa* species, concluded that there are positive yield and productivity responses to some degree of shade. For example, radiation use efficiency is not constant but increases at lower light flux densities. Under full light, light saturation density of *Musa* leaves is often exceeded and photorespiration

can occur (Eckstein et al. 1997). However, if shade exceeded around 30%, the planting-to-harvest period was prolonged and growth and yield reduced. Yet the majority of these studies were performed on Cavendish bananas, and data on the responses of plantain, cooking banana and other cultivars of banana are scarce.

In our trial, there were negative correlations at 5 MAP between growth variables and light level, with highest growth occurring under the lowest light intensity. This contrasts with data from Hauser (2010) in the same location where FHIA-21 plantains were grown in rows alongside *Inga edulis* Mart. shade trees. He found negative correlations at 3 MAP between canopy cover of the shade tree and growth of FHIA-21, but the soils had much higher Mg and K concentrations (2.35 and 0.37 cmol(+) kg<sup>-1</sup> for Mg<sup>2+</sup> and K<sup>+</sup> at 0–10 cm depth and 1.73 and 0.26 cmol(+) kg<sup>-1</sup> for Mg<sup>2+</sup> and K<sup>+</sup> at 10–20 cm) than the soils

used here (average of 1.61 and 0.14 26 cmol(+) kg<sup>-1</sup> for Mg<sup>2+</sup> and K<sup>+</sup> at 0–15 cm depth), so light may have limited nutrient uptake on the higher fertility soils. Thus, although in our trial the leaf and dry matter production were significantly higher under the heavily shaded (33% light) treatment, this may not occur on higher fertility soils where growth may be more limited by light level and less constrained by soil factors.

Given the importance of plantain as a smallholder crop, the major impact of BLS D as a production constraint, and the difficulty in managing it, surprisingly few investigations into the effects of shade on BLS D have been undertaken. It has been hypothesised that the phytotoxins of BLS D are activated by light (Daub and Hangarter 1983). However, recent work (Cruz-Cruz et al. 2011) did not find any light dependence of the activity of the BLS D phytotoxins. Germination of the BLS D infective agents requires high humidity and early growth is influenced by temperature (e.g. Jacome et al. 1991). Shading reduces air temperature below the optimum temperature therefore infection may be reduced. In the current trial, there was a clear reduction in BLS D attack under shade, however, the two shade treatments (67% light and 33% light) could not be distinguished.

Clearly, plantain growth was limited on grassland soils, but we are unable to attribute the growth limitation to the chemical variables measured. Other factors, possibly microbiological, might play a role in limiting growth on grassland soils and this needs to be investigated. In a study comparing soil enzymatic activity in forest soil to grassland soil excavated from the same area, beta-glucosidase activity was significantly different and approximately three times higher in the forest compared with grassland soil (B. Wick and co-workers, unpublished) and such shifts in enzymatic activity indicate changes in microbial community structure (Waldrop et al. 2000). For example, in a pot experiment, Tsané et al. (2005) demonstrated that the biomass of plantain grown on nutrient-poor soils – which were similar in properties to the ones used here – approximately tripled when plants were inoculated with mycorrhizal fungi. While there are few references comparing mycorrhizal fungi populations in grassland and forest soils in the tropics, a study in Central America showed that diversity, although not densities, tended to be higher in forest than in grassland, although patterns were not consistent (Johnson and Wedin 2007). In our experiment, possibly mycorrhizal fungi associations were formed in the forest-derived soil but not in the grassland-derived soil.

If the treatment of 67% shade on forest soil is defined as current farming practice, both removing shade and moving the plantings to grassland soil result in growth reductions regardless of cultivar. As FHIA-21 performed better than Batard in these experiments, growth reductions could be limited, but not eliminated,

by introducing this cultivar. If cultivar introduction were combined with a medium level of shading, such as 67% light level, possibly by using suitable shade trees, BLS D damage would be further limited. More research is needed on the factors limiting growth of plantains on grassland soils.

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