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GENOTYPIC DIFFERENCES IN THE GROWTH OF BANANAS (*MUSA* SPP.) INFECTED WITH MIGRATORY ENDOPARASITIC NEMATODES. 2. SHOOTS

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SUMMARY

In order to identify the effect of burrowing nematodes on the shoots (pseudostem and leaves) of banana plants and to determine whether or not shoot characteristics are associated with plant resistance to nematodes two experiments were conducted in controlled conditions within polytunnels. The banana plants were harvested on three occasions for the measurement of root morphology and biomass. Varieties differed in their resistance to nematodes from resistant (Yg Km5, FHIA 17, FHIA 03) and partly resistant (FHIA 01, FHIA 25) to not resistant (FHIA 23, Williams). Nematodes reduced total plant dry weight at the first harvest in Experiment 1 and by an average of 8.8 % in Experiment 2, but did not affect leaf area in either experiment. The ratio of above-ground weight to total plant weight was reduced from 75 % to 72 % in nematode-infected plants compared with the control plants for all varieties tested in Experiment 1, but was only reduced in FHIA 25 and FHIA 23 in Experiment 2. Varieties differed in above-ground growth. The FHIA varieties had greater shoot weights and leaf area than YgKm5 and Williams. Overall, resistance to nematodes was associated with the partitioning of a greater proportion of biomass to the roots than to above-ground parts

INTRODUCTION

Pests and diseases are a substantial threat both to commercial banana production for international trade, and for subsistence production and local consumption (Jeger *et al.*, 1995). Plant parasitic nematodes are among the main constraints to banana production. Although a number of nematode species infect bananas, the burrowing and lesion nematodes (*Radopholus similis* and *Pratylenchus* spp.) are considered to be the main problem in intensive commercial banana production, especially in the Cavendish group (Gowen and Quénéhervé, 1990; Sarah, 1989).

The widespread occurrence of nematodes and the large resultant yield losses have encouraged genetic improvement for resistance in banana. Plant resistance is an important component in the effective nematode management required for efficient crop production in the presence of nematode population densities that exceed the damage threshold (Starr *et al.*, 2002). Resistance of plants to nematodes is defined as the ability of the plant to suppress nematode reproduction (Bos and Parlevliet, 1995). Tolerance describes the sensitivity of a host to parasitism or the amount of damage

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sustained, and is measured ideally in terms of yield suppression (Cook and Evans, 1987; Hussey and Boerma, 1981).

Screening among bananas and plantains for resistance and tolerance to nematodes aims to identify genotypes with acceptable agronomic characteristics and a favourable response to nematodes for use in breeding programmes. Screening studies have shown different responses by banana plants to nematodes. For example, Mateille (1993) reported that the total leaf surface of nematode-infected plants was reduced more on the susceptible variety Poyo than on the resistant Gros Michel. However, selected *R. similis* populations had no effect on the percentage dry matter, chlorophyll content and nutrient assimilation of either variety. Hahn *et al.* (1996) reported that nematodes reduced the shoot dry weight of the susceptible variety Poyo but had no effect on the shoot weight of the resistant variety YgKm5. Barekye *et al.* (2000) reported that *R. similis* and *Helicotylenchus multicinctus* did not affect the height, girth and number of functional leaves of bananas. Guedira *et al.* (2004) reported that inoculation with *R. similis* reduced the length and diameter of the pseudostem as well as above-ground biomass in both the resistant and susceptible genotypes that they tested. Therefore, different plant responses to nematode infection exist within current banana germplasm.

Kalorizou *et al.* (2007), in a companion paper, reported that nematodes reduced the root dry weight and the number of primary roots of some banana varieties, thus representing a range of degree of resistance to nematodes. Those varieties that were resistant to nematodes had a greater root mass, and more and larger primary roots than non-resistant varieties.

The objective of this complementary study was to examine the effect of nematodes on the above-ground growth of varieties of bananas differing in their degree of resistance to nematodes, and to identify shoot characteristics associated with this resistance.

MATERIAL AND METHODS

Two experiments were conducted within controlled polytunnels at Reading, UK. These are fully described in Kalorizou *et al.* (2007). Seven varieties of banana (*Musa* spp.) were compared: Yangambi Km5, FHIA 17, FHIA 03 (resistant to nematodes); FHIA 01, FHIA 25 (partially resistant) and FHIA 23, Cavendish Williams (not resistant). Experiment 1 was a factorial design of four blocks, each with two nematode levels (with and without nematodes), three harvest times (202, 236, 270 days after planting, DAP) and five varieties (Yg Km5, FHIA 01, FHIA 03, FHIA 17 and FHIA 23). Twelve plants of each variety growing in 1-litre pots in a glasshouse were inoculated with 4500 nematodes comprising a mixture of *R. similis, Pratylenchus goodeyi*, *P. coffeae* and *H. multicinctus* and 12 plants were not inoculated (the control plants).

Fifteen days after inoculation the plants were moved to a polytunnel with variable temperature (mean = 20.6 °C, range 15.9–27.2 °C) and natural daylength, and were transplanted into 5-litre, then 15-litre pots containing vermiculite, sand and gravel (in the ratio 4:1:1). They were watered and fertilized by incorporation of Multicote 4 controlled-release fertilizer (Haifa Chemicals Ltd, Israel) into the mix at 119 DAP and using a complete nutrient solution (N-14, P-10, K-27, MgO-2.5 and SO_3–11) daily from 202 DAP.

Experiment 2 was a randomized block design of five blocks each with a factorial combination of two nematode levels (with and without), six varieties (Williams, Yangambi Km5, FHIA 01, FHIA 17, FHIA 23 and FHIA 25) and three harvest times (290, 320 and 350 DAP). Fifteen plants per variety growing in 1-litre pots were inoculated with 1750 nematodes (a mixture of *Pratylenchus goodeyi* and *R. similis*) and the same number of plants per variety was used as the control. The plants were moved to a polytunnel and re-potted twice into 5-litre, then 25-litre pots at about monthly intervals into a mixture of vermiculite, gravel and sand (4:2:1). Experiment 2 was conducted in a polytunnel with a constant air temperature of 29 °C/25 °C day/night until 320 DAP when the temperature was reduced to 25 °C/20 °C, a 12 hour thermoperiod, an average 74 % relative humidity, natural sunlight and daylength. Plants were irrigated daily with a complete nutrient solution (Summerfield *et al.*, 1977).

Data collection

At each harvest in Experiment 1, the dry weights of the above-ground parts of the mother plant and the roots were measured by sampling about 10 % of the fresh weight and drying at 80 °C until a constant weight was reached. In Experiment 2, the dry weight of above-ground parts of the mother plant (AGDW), the above-ground parts of the suckers and the roots were measured. Thus, total plant dry weight (TDW) was AGDW plus roots in Experiment 1, and AGDW plus roots plus sucker dry weight in Experiment 2. The green leaf area of each plant was measured in each experiment using a Delta T leaf area scanning machine.

Statistical analysis

Data were analysed using the SPSS statistical package (SPSS, 2001). Analysis of variance was used to assess treatment effects. The root and shoot dry weights and leaf area data were transformed to $\log_{10}$ (x) before analysis to ensure homogeneity of residuals. Mean separation was made using the standard error of differences at the 5 % level when significant differences among treatments were found. In each experiment, the linear regression between resistance to nematodes (calculated as the number of nematodes for each variety as a proportion of the number of nematodes on the variety which had the fewest (Table 2 of Kalorizou *et al.* (2007)) and reduction in shoot dry weight due to nematodes (as a proportion of the dry weight of the control) was examined.

RESULTS

Plant biomass

Total plant dry weight (TDW) in Experiment 1 was affected by the interaction between variety, nematode and time ($p = 0.009$). At 202 DAP, nematodes reduced the TDW of FHIA 01, FHIA 03 and FHIA 23 compared with the control plants (Figure 1). Nematodes reduced TDW of all varieties except FHIA 23 at 236 DAP, but

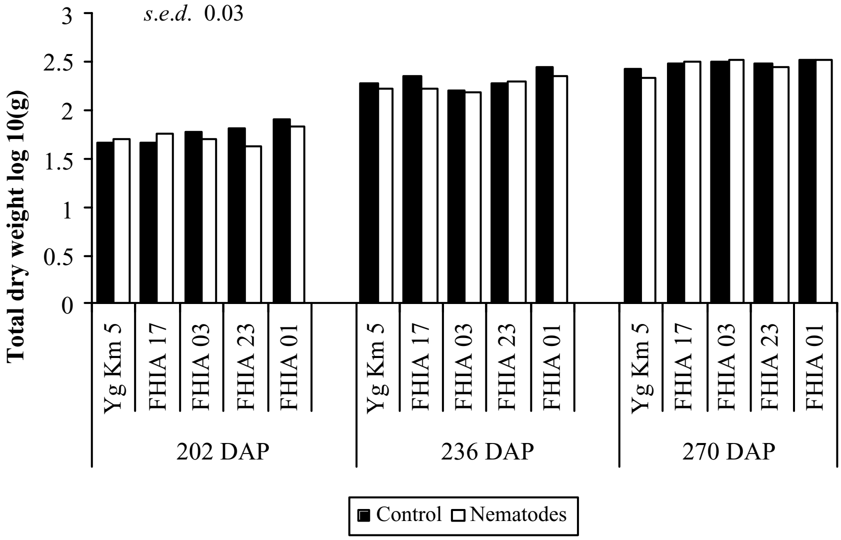

Figure 1. Effect of variety and nematode on total dry weight at three harvests in Experiment 1.

at 270 DAP nematode-infected and control plants had the same TDW. The effect of the interaction between variety and time on TDW was also significant ($p < 0.001$). Of all varieties tested at 202 and 236 DAP, FHIA 01 had the greatest TDW, but values were similar to FHIA 03 and FHIA 17 at 270 DAP (Figure 1).

In Experiment 2, the effects of the interaction between variety, nematode and time on TDW were not significant ($p = 0.857$), but the varieties differed ($p < 0.001$). FHIA 23 had the lowest TDW and FHIA 25 the greatest (data not shown). Nematodes reduced TDW at 350 DAP ($\log_{10}$ (g) = 3.1 for control plants and $\log_{10}$ (g) = 3.06 for nematode-infected plants, *s.e.d.* = 0.016).

The interaction between variety, nematode and time affected AGDW in Experiment 1 ($p = 0.026$). FHIA 17 and YgKm5 nematode-infected plants had a greater AGDW than the control plants at 202 DAP (Figure 2). At 236 DAP, nematodes reduced the AGDW of all varieties, but at 270 DAP nematode-infected and control plants had the same AGDW. On average, nematodes reduced the AGDW (mean value for control and nematode-infected plants $\log_{10}$ (g) = 2.05 and $\log_{10}$ (g) = 2.0, respectively, *s.e.d.* 0.01).

The AGDW was affected by the interaction between variety and time ($p < 0.001$). On each occasion FHIA 01 had the greatest AGDW (Figure 2). At 202 and 270 DAP, YgKm5 had the smallest AGDW, while FHIA 03 had the smallest AGDW at 236 DAP. There was no linear correlation between the relative number of nematodes per root system and the percentage reduction in AGDW caused by nematodes ($p = 0.706$).

In Experiment 2 there was no effect of the interaction between variety, nematode and time on the AGDW ($p = 0.739$). Varieties differed in AGDW and were subject to an interaction between variety and time ($p = 0.046$). At 290 DAP FHIA 25 and FHIA 17 had a greater AGDW than the other varieties (Figure 3). At 320 and 350

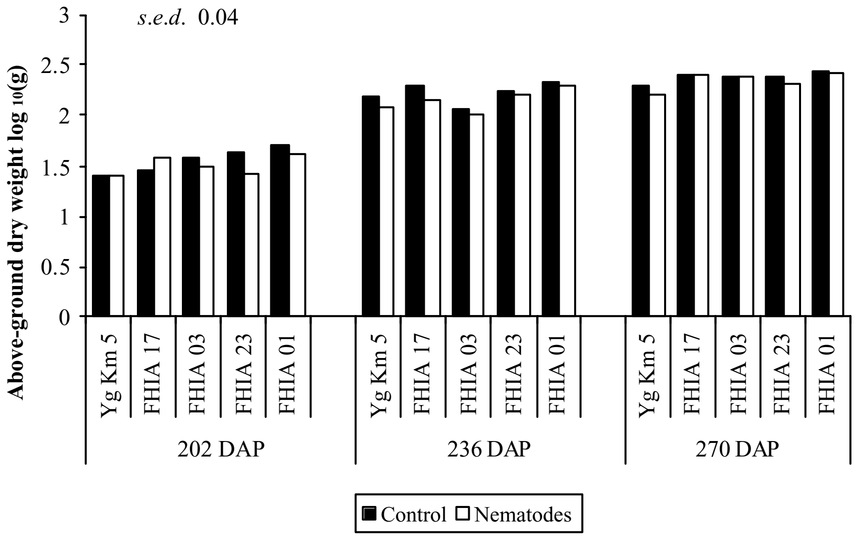

Figure 2. Effect of variety and nematode on the above-ground dry weight at three harvests in Experiment 1.

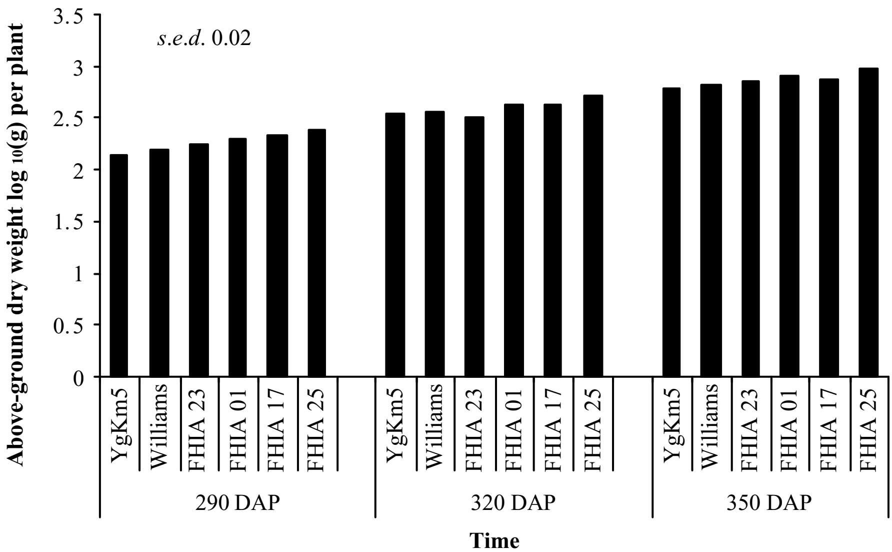

Figure 3. Above-ground dry weight of banana varieties at three harvests in Experiment 2 expressed as the mean of the control and nematode-infected plants.

DAP, FHIA 25 had the greatest AGDW and YgKm5 and Williams had the smallest AGDW at 320 DAP.

The interaction between variety and nematode affected AGDW ($P = 0.018$). FHIA 23 and FHIA 25 nematode-infected plants had smaller AGDW than the control plants (mean values for control and nematode-infected plants were $\log_{10}$ (g) = 2.58 and $\log_{10}$ (g) = 2.49, respectively, for FHIA 23 and $\log_{10}$ (g) = 2.72 and $\log_{10}$ (g) = 2.68 for

FHIA 25, *s.e.d.* = 0.018). Nematodes did not affect the AGDW of the other varieties. There was no linear correlation between the relative number of nematodes per root system and the percentage reduction in the AGDW caused by nematodes ($p = 0.706$).

Leaf area

There was no evidence of any interaction affecting leaf area in Experiment 1 and 2, but varieties differed ($p = 0.011$ and $p < 0.001$ for Experiments 1 and 2, respectively). In Experiment 1, FHIA 01 had a greater leaf area than FHIA 17 and YgKm5 (0.79 m^2, 0.72 m^2, and 0.67 m^2 respectively). All the other varieties had similar leaf areas. In Experiment 2, YgKm5 had the smallest leaf area (0.43 m^2) and FHIA 25 the greatest (0.65 m^2). Nematodes did not affect leaf area ($p = 0.479$ and $p = 0.053$ for Experiments 1 and 2, respectively).

Dry matter distribution

In Experiment 1, the effect of the interaction between variety, nematode and time on the ratio of the above-ground dry weight to total dry weight (AGDW:TDW) was not significant ($p = 0.189$). However, the interaction between variety and time affected this ratio ($p = 0.009$). At 202 DAP, YgKm5 had the smallest ratio (Figure 4). At 236 and 270 DAP, FHIA 03 and YgKm5 had the smallest ratio. All the other varieties had the same AGDW:TDW ratios. This was greatest at 236 DAP. Nematodes reduced the ratio (mean value for control and nematode-infected plants 0.751 and 0.723, respectively, *s.e.d.* = 0.01).

In Experiment 2, the effect of the interaction between variety, nematode and time on AGDW:TDW ratio was not significant ($p = 0.671$). There was evidence of an effect

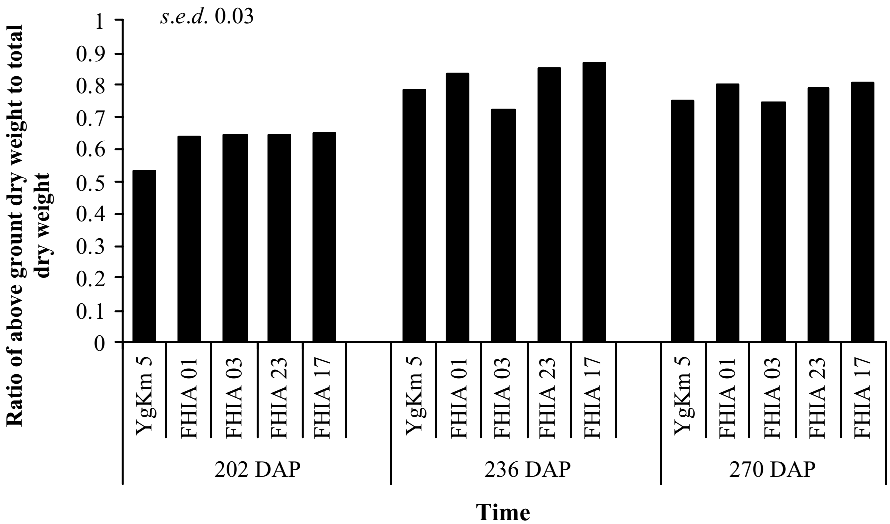

Figure 4. The ratio of the above-ground to total dry weight of banana varieties at three harvests in Experiment 1 expressed as the mean of the control and nematode-infected plants.

of the interaction between variety and nematode on this ratio ($p = 0.016$), FHIA 23 and FHIA 25 nematode-infected plants having a smaller ratio than the control plants (Figure 5). In contrast, nematode-infected plants of FHIA 01 and YgKm5 had a greater ratio than controls. The interaction between variety and time also affected the AGDW:TDW ratio ($p = 0.002$). At 290 DAP, YgKm5 had the smallest ratio (Figure 6).

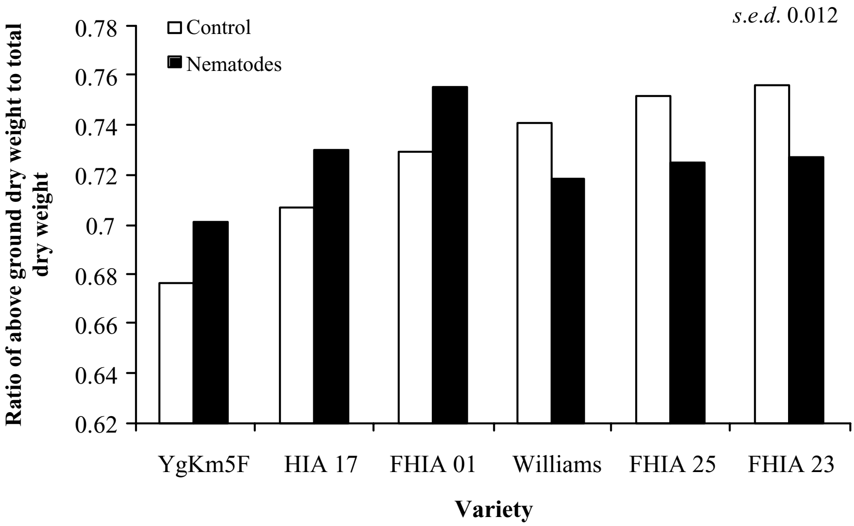

Figure 5. Effect of nematodes on the ratio of the above-ground to total dry weight of banana varieties in Experiment 2.

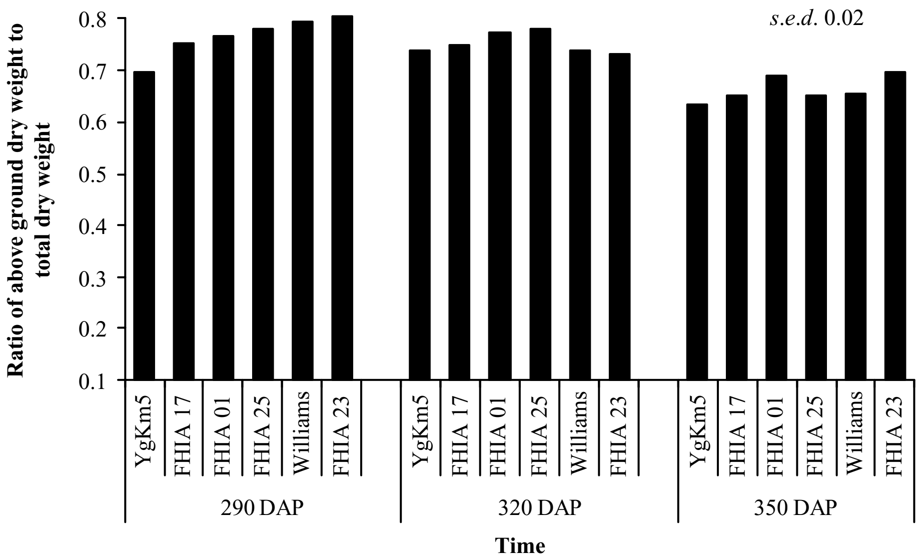

Figure 6. The ratio of the above-ground to total dry weight of banana varieties at three harvests in Experiment 2 expressed as the mean of the control and nematode-infected plants.

At 320 DAP, all the varieties had the same ratio, but at 350 DAP, FHIA 23 and FHIA 01 had the greatest ratio. AGDW:TDW decreased over time.

In Experiment 1, there was no evidence of any interaction on the ratio of leaf dry weight to total dry weight (LDW:TDW). This ratio also decreased over time, but was the same for all varieties ($p = 0.221$) and was not affected by nematodes ($p = 0.579$). In Experiment 2, the effect of the interaction between variety, nematode and time on LDW:TDW was not significant ($p = 0.999$). The only evidence of an interaction was between variety and nematode ($p = 0.039$). FHIA 01 nematode-infected plants had a greater ratio than the control plants (mean value for control and nematode-infected plants 0.73 and 0.76, respectively, *s.e.d.* = 0.012). In contrast YgKm5, Williams, FHIA 23 and FHIA 17 nematode-infected plants had the same ratio as the control plants. The LDW:TDW ratio differed among varieties ($p < 0.001$). Williams and YgKm5 had the greatest and smallest ratios on average, respectively. These ratios increased until 320 DAP but decreased thereafter.

DISCUSSION

The direct effect of nematodes on plants is on the roots where the destruction of root and corm tissues reduces water and mineral uptake and results in reduced growth and development (Sarah *et al.*, 1996). Root system development and shoot growth in banana are highly related (Pearsall, 1927). Consequently, a reduction in root growth due to nematode infection might be associated with a reduction in growth above-ground. In Experiment 1, nematodes reduced the total and above-ground dry weights until 202 DAP. In Experiment 2, nematodes reduced the above-ground dry weight of FHIA 23 and FHIA 25 and the total dry weight of all the varieties tested. Infection of banana roots nematodes therefore reduced the biomass accumulation of the above-ground parts of the plant.

Nematodes did not affect the leaf area of any variety. This observation agrees with the results of Barekye *et al.* (2000) who reported that nematodes did not affect the number of functional leaves of banana. In both our experiments, non-resistant and partially resistant varieties had a greater leaf area than the resistant varieties. However, for some varieties, with more nematodes on the roots (Table 2, Kalorizou *et al.*, 2007), such as FHIA 01 and Williams, the above-ground parts were not affected by nematodes. Perhaps plants with large leaf areas (Figure 6), and thus greater photosynthetic capacity, can partly mask nematode damage to aerial parts. The small reduction in the above-ground dry weight due to nematode infection found in both experiments indicates that *Musa* spp. plants can grow vigorously even when infected.

Dry matter partitioning between the root and the shoot varied between genotypes and with the developmental stage of the plant. The shoot:root ratio represents a balance between shoot and root activity (Davidson, 1969), which is affected by environmental conditions. In this study, plants invested a greater proportion of biomass in the above-ground parts than in the roots; the ratio of above-ground to total dry weight ranged between 0.55 and 0.80. A previous estimate of this for bananas growing at 25/18°C was towards the upper limit of this range at 0.78 (Turner and Lahav, 1983). Differences in the dry matter partitioning to roots has been observed between the resistant variety Gros Michel (AAA) and the susceptible variety of the Cavendish group Grande Naine (AAA) (Stover and Buddenhagen, 1986). In our study there was an association between dry matter distribution and resistance to nematodes. For example, the partially resistant variety FHIA 01 and the non-resistant variety FHIA 23 had greater above-ground to total dry weight ratios than the resistant varieties YgKm5 and FHIA 03. Thus, resistance to nematodes was associated with varieties that apportioned comparatively more biomass to the roots than to the above-ground parts.

Mateille (1993) proposed that the leaf length:blade area ratio could be used as an agronomic screening parameter for banana resistance to *R. similis*. In the current study, nematodes did not affect the leaf area. However, some of the partially resistant and non-resistant varieties had the largest leaf area and the resistant variety Yg Km5 had the smallest in both experiments.

According to Hahn *et al.* (1996), the reproductive fitness of burrowing nematode populations is not necessarily associated with significant damage to banana. Nematode density is, however, the most appropriate variable to use to look at the relationship between growth reduction and nematode pathogenicity (Hahn *et al.*, 1996), but no linear correlation between the relative density of nematodes on the roots and the decrease in above-ground dry weight was found in this study. Therefore, there was no simple relationship between resistance and tolerance to nematodes for above-ground (this study) and below-ground (Kalorizou *et al.*, 2007) growth.

Banana and plantain (*Musa* spp.) are important cash and subsistence crops in many tropical and subtropical regions of the world (Robinson, 1996). Because of biotic stresses, considerable efforts have been made to increase resistance to pests and diseases and to broaden the genetic base of the crop (Vuylsteke *et al.*, 1997). Blomme *et al.* (2001) reported that diploids (*Musa* AA) and dessert bananas (*Musa* AAA) had small values for most root and shoot growth characteristics, while plantains (*Musa* AAB), cooking bananas (*Musa* ABB) and tetraploids (*Musa* AAAB) had larger values. Genotypic differences in root and shoot growth were observed among the varieties studied here and reported in Kalorizou *et al.* (2007), and some of these can be associated with differences in resistance to burrowing nematodes. Resistant varieties partitioned a greater proportion of biomass to their roots that contributed to a greater root mass, and more primary roots, than less resistant varieties. Information such as this should aid the identification of plant characteristics for banana genotypes that are more resistant and/or tolerant to burrowing nematodes.

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