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Field trial evaluation of nematode susceptibility within *Musa*

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Summary – Fifty-two clones of *Musa* were evaluated for their susceptibility to the plant-parasitic nematodes *Radopholus similis*, *Hoplolaimus pararobustus* and *Meloidogyne incognita* in two field trials. As a group the AAB Plantains showed the greatest susceptibility to both *R. similis* and *H. pararobustus*. *Musa* AAA and ABB types tended to show lower susceptibility to *R. similis* and some types exhibited near resistance. *H. pararobustus* and *M. incognita* occurred in lower numbers than *R. similis* and intervarietal differences in susceptibility were less marked.

Résumé – ***Évaluation par essais en champ de la sensibilité aux nématodes chez le genre* Musa** – Au cours de deux essais en champ, cinquante-deux espèces et variétés de *Musa* on été testées pour leur sensibilité envers les nématodes *Radopholus similis*, *Hoplolaimus pararobustus* et *Meloidogyne incognita*. Le groupe des bananiers plantains AAB présente la plus grande sensibilité tant envers *R. similis* que *H. pararobustus*. Les types AAA et ABB marquent une tendance vers une moindre sensibilité à *R. similis*, quelques variétés étant même proches de la résistance. *H. pararobustus* et *M. incognita* sont moins représentés que *R. similis* et dans leur cas les différences variétales de sensibilité sont moins marquées.

Key-words : *Musa*, plantains, bananas, nematodes, *Radopholus similis*, resistance, susceptibility.

Of the plant-parasitic nematodes known to cause damage to bananas and plantains, *Radopholus similis*, is considered the most important (Gowen & Quénéhervé, 1990) in particular due to its status as the most important nematode pest of commercial plantations. In Africa this nematode also poses a major and increasing threat to smallholder agriculture (Sarah, 1989). With increased awareness of the important role bananas and plantains occupy in domestic consumption there is a need for a better understanding of the host parasite relationships between plant-parasitic nematodes (in particular *R. similis*) and *Musa* varieties other than the AAA Cavendish cultivars upon which the commercial sector relies (Anon., 1988). Such information is needed both because breeders have so far successfully transmitted resistance to *R. similis* from only source (Pinochet, 1988 *a*) and because of suggestions that known naturally occurring cultivars with greater disease resistance may be promoted until plant breeding is able to provide improved varieties (Champion, 1976). Following a reorganization of the *Musa* germplasm collection at the Institut de la Recherche Agronomique Njombe Station, Littoral Province, Cameroon, planting material comprising 52 clones representing various genetic groups of *Musa* became available and was used in two field trials.

Materials and methods

Two adjacent trials (A and B) identical in design were planted in April 1989. The site is in the humid lowland tropical zone of the country 80 m above sea level in a region of leached sandy soils of volcanic origin. Soils tend to be acid (pH 5.1-6.1) and deficient in potassium (Delvaux, 1989). One month prior to planting these trials the land had been growing the AAA Cavendish banana variety Poyo and was known to be infested with *R. similis*.

Planting material (trimmed " suckers ") was planted in rows 3.2 m × 1.5 m. Each trial comprised 27 different clones and both included the two AAA Cavendish bananas Poyo and Grande Naine as reference varieties. Both trials were in randomized block designs of eight replicates, each plot consisting of a single plant, each variety representing a treatment and surrounded by a single outside border of Poyo. No nematicides or insecticides were applied, aerial spraying against Black Sigatoka (*Mycosphaerella fijiensis* var. *difformis* Mulder & Stover) occurred as necessary. Fertilization followed normal procedures and a limited amount of irrigation took place during the dry season.

Trials were sampled in September and October 1990. Root sampling and nematode extraction followed established procedures (Quénéhervé & Cadet, 1986; Pinochet, 1988 *b*). Each plant provided one sample, 25 g of root material being washed, blended and passed through screens of 250 μm, 50 μm and 32 μm apertures. Material retained by the 250 μm screen was discarded, material collected on the other screens was made up to 100 ml with water to form the sample. In cases of very high nematode numbers the sample was diluted to 500 ml. Nematodes in three 1-ml aliquots from each sample were counted. A complete block was processed in a single day, blocks being sampled sequentially on consecutive days.

Counts of the three most commonly found plant-parasitic nematode species are reported here as numbers of nematodes per 100 g root fresh weight (RFW). Numbers of *R. similis* consist of adult females plus immature stages (ie. excluding adult males). Numbers of *Meloidogyne incognita* consist of all endoparasitic stages plus adult males but does not include vermiform second-stage juveniles. All *Hoplolaimus pararobustus* were counted.

After the log-transformation *($log_e (n + 1)$)* results were subjected to Analysis of Variance using GENSTAT (Genstat, 1987) and the Duncans' Multiple Range Test was applied.

Results

Details of *Musa* clones used and nematode numbers obtained for trials A and B are shown in Tables 1 and 2, respectively.

Greater numbers of *R. similis* occurred than of the other two nematode species. Maximum observed values were 299 330 *R. similis* per 100 g RFW for the AAB Plantain Batard in Trail A and 296 670 *R. similis* per 100 g RFW for the AAB Plantain Obel in Trial B. Highest mean values (de-transformed from the $log_e (n + 1)$ means) were 32 859 *R. similis* per 100 g RFW for the AAB variety Laknao and 13 359 *R. similis* per 100 g RFW for the AAB Plantain Esang for trials A and B respectively (log-transformed means 10.4 ± 0.7 and 9.5 ± 0.7 respectively). The least susceptible variety in Trial A was the AAA Ibota group banana Yangambi with 4.0 *R. similis* per 100 g RFW (log-transformed mean 1.6 ± 0.7), and in Trial B the AAB Pisang Kelat with 6.0 *R. similis* per 100 g RFW (log-transformed mean 1.9 ± 0.7).

The maximum observed numbers of *H. pararobustus* found in trial A was 14 000 per 100 g RFW on *M. acuminata* and in trial B 18 670 per 100 g RFW on the AAB Plantain Obel. Maximum means (de-transformed from the $log_e (n + 1)$ means) were 298.0 on the AAA Cavendish Grande Naine in Trial A and 3293.0 on the AA Pisang Trimulin in Trial B (log-transformed means, 5.7 ± 0.6 and 8.1 ± 0.7 g RFW, respectively). *H. pararobustus* numbers were generally low and few clones differed significantly in susceptibility.

Root-knot nematode, *Meloidogyne incognita*, was the least abundant nematode. Maximum observed numbers were 10 000 per 100 g RFW on the AAB Plantain French Sombre in trial A and 9330 per 100 g RFW on the ABB Bluggoe Christine in trial B. Maximum means (de-transformed from the *($log_e (n + 1)$* means) were 98.0 *Meloidogyne incognita* per 100 g RFW on *Musa basjoo* in trial A and 89.0 *Meloidogyne incognita* per 100 g RFW on *Musa acuminata burmannica* in Trial B (log-transformed means 4.6 ± 0.6 and 4.5 ± 0.6 respectively). As with *H. pararobustus* inter-clonal differences were not market. No *Meloidogyne incognita* were recorded on the AAB variety Laknao although vermiform J2 s were observed in some samples.

Discussion

Total resistance (immunity) was not confirmed for any of the three nematode species considered and, as in other work, only differences between clones in susceptibility were observed (Gowen, 1976; Pinochet, 1988 *a*). The predominant nematode species present was *R. similis,* as found in Cavendish banana plantations elsewhere in West Africa (Quénéhervé *et al.,* 1991). *Helicotylenchus multicinctus* which can outnumber *R. similis* in some areas (Gowen, 1979) was found infrequently and was not evaluated.

These results show a high level of susceptibility to *R. similis* among the AAB Plantains. In both trials, six of the ten most susceptible clones were AAB Plantains (i.e. twelve out of the total of seventeen AAB Plantain varieties evaluated) and the closely related AAB variety Laknao was the most susceptible clone in Trial A. This is believed to be the first extensive evaluation of varietal susceptibility conducted in Africa and is clear evidence of the threat this nematode poses to this important crop (Sarah, 1989).

By contrast the AAA Cavendish varieties, mainstay of the commercial (banana export) industry, on which much money and effort is spent in chemical control (Gowen & Quénéhervé, 1990) were more resistant than the AAB Plantains. These results agree with those of Perez *et al.* (1986) in Cuba, reporting greater susceptibility, to both *R. similis* and *Pratylenchus coffeae,* of AAB Plantains compared with AAA Bananas. Maximum *R. similis* numbers observed on the reference variety Grande Naine, 86 000 and 56 000 *R. similis* per 100 g root for Trials A and B respectively, were approximately a quarter of the maxima recorded on AAB Plantains. Means numbers of *R. similis* found in the reference varieties (Tables 1 and 2), correspond well with field populations reported on Cavendish bananas elsewhere (Sarah & Vilardebó, 1979; Davide, 1980; Loridat, 1989).

Bananas of the Gros Michel group have long been considered less susceptible to *R. similis* than Cavendish varieties (Simmonds, 1966). Using a root-lesion index Wehunt *et al.* (1978) found roots of the Gros Michel var. Cocos to be significantly less damaged than roots of the Cavendish varieties they evaluated. Davide and Marasigan (1985) also reported lower numbers of *R. similis* in roots of Gros Michel than Cavendish and Trial A (Table 1) showed Gros Michel to be significantly less susceptible than two of the three Cavendish varieties included. Mateille (1992), in a study using tissue cultured plants, attributes this lower susceptibility of Gros Michel (compared to the Cavendish variety Poyo) to

Table 1. *Numbers[1] of* Radopholus similis[2], Hoplolaimus pararobustus *and* Meloidogyne incognita[3] *per 100 g Fresh Weight Root on 27 field grown* Musa *clones. Njombe Field Trial A.*

Clone	Genome	Genetic Group	*R. similis*		*H. pararobustus*		*M. incognita*	
			Numbers	DMR	Numbers	DMR	Numbers	DMR
Laknao	AAB	Laknao	32 858.6	*a*	4.5	*e*	0.0	*e*
Ebanga	AAB	Plantain	24 342.0	*ab*	39.4	*abcdef*	1.5	*cde*
French Clair	AAB	Plantain	24 342.0	*ab*	23.5	*cdefg*	5.0	*bcde*
Batard	AAB	Plantain	13 358.0	*abc*	11.2	*cdefg*	32.1	*ab*
Mujuba	AAA	Lujugira	10 937.0	*abcd*	2.0	*gh*	5.0	*bcde*
Corne N° 5	AAB	Plantain	10 937.0	*abcd*	243.7	*ab*	43.7	*ab*
Popoulou	AAB	Popoulou	8 102.1	*abcd*	35.6	*abcdef*	13.9	*abcd*
French Sombre	AAB	Plantain	6 634.2	*abcd*	35.6	*abcdef*	2.0	*cde*
Corn Type	AAB	Plantain	6 001.9	*abcd*	11.2	*cdefgh*	1.2	*de*
Grande Naine	AAA	Cavendish	4 446.1	*abcd*	297.9	*a*	11.2	*abcd*
Musa basjoo			3 639.9	*abcde*	19.1	*cdefgh*	98.5	*a*
Musa acuminata Type II			2 996.3	*bcde*	17.2	*cdefgh*	3.9	*bcde*
Bluggoe	ABB	Bluggoe	2 439.6	*cdef*	3.9	*fgh*	1.2	*de*
Figue Pomme Adju	AAA	Lujugira	1 635.0	*cdef*	4.5	*efgh*	1.0	*de*
Madre del Platano	AAB	Plantain	1 479.3	*cdef*	243.7	*ab*	10.0	*bcd*
Pisang Mas	AA		1 211.0	*defg*	5.0	*efgh*	17.2	*abcd*
Poyo	AAA	Cavendish	1 095.6	*defg*	10.0	*defgh*	4.5	*bcde*
Americani	AAA	Cavendish	445.9	*efgh*	39.4	*abcdef*	5.0	*abc*
Musa textilis[4]			269.4	*fgh*	48.4	*fgh*	5.0	*bcde*
Pelipita	ABB	Pelipita	163.0	*ghi*	3.9	*fgh*	11.2	*abcd*
Musa coccinea			108.9	*hi*	29.0	*bcdefg*	0.0	*e*
Musa balbisiana (CMR)			98.5	*hi*	1.5	*h*	4.5	*bcde*
Gros Michel	AAA	Gros Michel	80.4	*hi*	65.7	*abcd*	1.2	*de*
Rajapuri India	AAB	Nendra Padaththi	53.6	*hi*	108.9	*abc*	5.0	*bcde*
1877	AAAA		48.4		48.4	*efgh*	12.5	*abcd*
Musa laterita			23.5	*ij*	6.4	*efgh*	6.4	*bcde*
Yangambi	AAA	Ibota	3.9	*j*	12.5	*cdefgh*	15.4	*abcd*

[1]Nematode numbers per 100 g Fresh Weight Root, de-transformed from the $log_e\ (n + 1)$ mean of 8 replicates; [2]*R. similis* adult females + juveniles; [3]*M. incognita* endoparasitic stages + males. [4]Possibly a *textilis* × *balbisiana* hybrid. For each nematode species the $log_e\ (n + 1)$ transformed means of *Musa* clones followed by the same letter do not differ significantly ($P = 0.05$) according to the Duncan's Multiple Range Test (DMR).

both reduced and delayed initial invasion and lower nematode multiplication.

H. pararobustus has been reported attacking both AAA Cavendish bananas and AAB plantains in Côte d'Ivoire in similar numbers to those reported here (Adiko, 1988; Mateille *et al.*, 1988). The AAB Plantains as a group (as with *R. similis*) again appear to be more susceptible than other types. Five of the seven AAB Plantains evaluated in Trial A and six of the ten evaluated in Trial B ranked among the ten most susceptible varieties in each trial. Two non-parametric tests failed to show statistically significant rank correlations between susceptibility to these two nematode species over all varieties evaluated. However, one may speculate that the greater susceptibility to both nematode species observed within the AAB Plantains may have a similar basis. Mateille (1992) found Gros Michel banana to be significantly more susceptible to *H. pararobustus* than the Cavendish variety Poyo. This was also seen in Trial A (Table 1) though the difference failed to reach statistical significance.

Root-knot nematodes are widely distributed and commonly found on bananas (Gowen & Quénéhervé, 1990) although their importance in causing yield loss may be more restricted, and complicated by possible interactions with other nematode species. Davide (1980) showed how *R. similis* numbers may increase at the expense of *M. incognita*. Santor and Davide (1982) described the histological incompatibility between the two species. A possible example of this is provided by the

AAB Laknao, rated root-knot susceptible by Davide and Marasignan (1985). In Trial A this variety was found free of *M. incognita* but with the highest population of *R. similis*. By contrast in Trial B the AAB Pisang Kelat was the variety most resistant to *R. similis* but had the second highest number of *M. incognita*. An antagonism of *R. similis* to *M. incognita* may explain the generally much lower numbers of *M. incognita* found in this work compared with numbers reported when using pure root-knot nematode populations (Davide & Marasigan, 1985). As found by these workers most varieties screened, including both *M. balbisiana* and *M. acuminata* showed some degree of susceptibility.

Most hopes for breeding resistance to *R. similis* are based on *Musa acuminata* varieties of the Pisang Jari Buaya (PJB) group and their progenies (Pinochet, 1988 *a*, 1992). Although no PJBs were included here some of the varieties tested appear, even allowing for possible differences in extraction efficiencies and population assessments, to show levels of *R. similis* resistance similar or greater to those reported for PJB and its progenies (Pinochet & Rowe, 1978). In Trial A the AAA Ibota group banana Yangambi showed resistance to *R. similis*, with a (de-transformed) mean population of six *R. similis* per 100 g RFW, an observation supported by greenhouse work (Sarah *et al.*, 1992). This variety appears to be resistant to another major pest of *Musa* in West Africa, the Banana Borer Weevil *Cosmopolites sordidus* Germar (Fogain & Price, 1993) and is also resistant to Black Sigatoka *Mycospharella fijiensis* var. *difformis* (Fouré, 1982). However both *M. acuminata* accessions used in this work, including the subspecies *burmannica* Type Calcutta 4 used in Trial B (the source of Black Sigatoka resistance in most breeding schemes) appear relatively susceptible to *R. similis* (Tables 1 and 2). In Trial B both the AB Safet Velchi and the AAB Pisang Kelat also appeared to support populations as low as reported on PJB and its progenies (Pinochet, 1992).

Table 2. *Numbers*[1] *of* Radopholus similis[2], Hoplolaimus pararobustus *and* Meloidogyne incognita[3] *per 100 g Fresh Weight Root on 27 field grown* Musa *clones. Njombe Field Trial B.*

Clone	Genome	Genetic Group	*R. similis*		*H. pararobustus*		*M. incognita*	
			Numbers	DMR	Numbers	DMR	Numbers	DMR
Esang	AAB	Plantain	13 358.7	*a*	811.4	*abcde*	89.0	*abcdef*
Obel	AAB	Plantain	7 331.0	*ab*	402.4	*abcde*	4.5	*bcdef*
Pisang Trimulin	AA		3 640.0	*abc*	3 293.5	*a*	11.2	*abcdef*
Mbouroukou N° 3	AAB	Plantain	3 640.0	*abc*	329.3	*bcdefg*	3.9	*cdef*
Kedong Kekang	AAB	Plantain	2 696.3	*abcd*	811.4	*abcd*	13.9	*abcdef*
Psi-Psi	AAB	Plantain	2 696.3	*abcd*	1 211.0	*abcd*	5.0	*bcdef*
Grande Naine	AAA	Cavendish	1 997.2	*abcd*	147.4	*cdefg*	5.0	*bcdef*
Mbotoko Rouge	AAB	Plantain	1 635.0	*abcd*	811.4	*abcd*	48.4	*ab*
Guyod	AA		1 635.0	*abcd*	444.9	*abcde*	13.9	*abcdef*
Williams	AAA	Cavendish	1 479.3	*abcd*	329.3	*bcdefg*	35.6	*abcd*
Figue Rose	AAA	Red	1 211.0	*abcde*	43.7	*fgh*	1.2	*ef*
M. acuminata burmannica Calcutta 4			991.3	*bcdef*	89.0	*defgh*	89.0	*a*
Cachao	ABB	Bluggoe	664.1	*cdef*	15.4	*h*	12.5	*abcdef*
1 Hand Planty	AAB	Plantain	543.6	*cdef*	89.0	*defgh*	43.7	*abc*
Plantain N° 17	AAB	Plantain	491.7	*cdefg*	297.9	*bcdefg*	1.2	*ef*
Plantain N° 2	AAB	Plantain	364.0	*cdefg*	89.0	*defgh*	17.2	*abcde*
Manneah	AAA	Cavendish	364.0	*cdefg*	147.4	*cdefg*	3.5	*def*
Poyo	AAA	Cavendish	243.7	*defgh*	133.3	*cdefgh*	6.4	*bcdef*
Christine	ABB	Bluggoe	120.5	*efghi*	402.4	*abcdef*	7.2	*abcde*
Thong Dok Mak	AA		98.5	*fghi*	243.7	*bcdefg*	17.2	*ef*
Big Ebanga	AAB	Plantain	48.4	*ghij*	1 095.6	*abc*	1.2	*ef*
Lacatan	AAA	Cavendish	26.1	*hij*	39.4	*gh*	8.2	*bcdef*
Foconah	AAB	Pome	26.1	*hij*	1 807.0	*ab*	1.0	*f*
Figue Pomme Ekona	AAB	Silk	21.2	*ij*	13.9	*h*	1.5	*ef*
Pisang Papan	AAA	Undetermined	15.4	*ij*	48.4	*fgh*	1.5	*ef*
Safet Velchi	AB		12.5	*ij*	80.4	*efgh*	5.7	*bcdef*
Pisang Kelat	AAB	Pisang Kelat	5.7	*j*	133.3	*cdefgh*	48.4	*ab*

[1]Nematode numbers per 100 g Fresh Weight Root, de-transformed from the $\log_e (n + 1)$ mean of 8 replicates; [2]*R. similis* adult females + juveniles; [3]*M. incognita* endoparasitic stages + males. For each nematode species the $\log_e (n + 1)$ transformed mean of *Musa* clones followed by the same letter do not differ significantly ($P = 0.05$) according to the Duncans Multiple Range Test (DMR).

Pinochet (1988 *a*) called for information on the nematode susceptibility of potential substitutes, among them the ABB variety Pelipita, for Black Sigatoka susceptible Plantain varieties. Davide and Marasignan (1985) rated Penipita *(sic)* as resistant to *R. similis.* This variety was significantly less susceptible to *R. similis* than any of the AAB Plantains evaluated in Trial A (Table 1). The (detransformed) mean number of *R. similis* (164 per 100 g root) on Pelipita was less than 1 % of the (detransformed) means recorded on the two most susceptible AAB Plantains in this trial. Although the AAB Plantains as a group exhibited high susceptibility to *R. similis* four other AABs, namely the varieties Foconah, Figue Pomme d'Ekona, Pisang Kelat and Rajapuri India of the Pome, Silk, Pisang Kelat and Nendra Padaththi Groups respectively, showed significantly lower susceptibility. Of the AAB Plantains evaluated the False Horn variety Big Ebanga showed similar low susceptibility to *R. similis,* an impression also gained during a field survey (Bridge *et al.,* unpubl.).

In conclusion this work demonstrates that a high degree of variability in susceptibility to *R. similis* exists within *Musa.* Of particular interest is how certain varieties are able to support much higher *R. similis* populations than other, the mechanisms behind this greater susceptibilty, and if such mechanisms are also associated with a degree of tolerance to this nematode. Although the search for resistance to *R. similis* has so far been disappointing (Pinochet, 1988 *a*) this work shows that mechanisms of much reduced susceptibility do exist. Reduced susceptibility could in itself be a major advance and this information may be of use, particularly with the possibilities offered by advanced breeding techniques (Stover & Buddenhagen, 1986).

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