"Yangambi km5" (*Musa* AAA, Ibota subgroup): a possible source of resistance to *Radopholus similis* and *Pratylenchus goodeyi*

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Summary - Field and shadehouse studies were carried out in Cameroon to compare the susceptibility to *Radopholus similis* of the "Yangambi km5" banana cultivar to that of other triploid *Musa* clones and also to evaluate its susceptibility to *Pratylenchus goodeyi*. Results of field trials have shown that populations of *R. similis* recovered from "Yangambi km5" every 2 months over a 2-year period were significantly lower than those from the other cultivars. A shadehouse experiment showed that different inoculum levels did not have any significant effect on the susceptibility of "Yangambi km5" to *R. similis*. Population levels of *R. similis* recovered 6 weeks after inoculation of "Yangambi km5" with 1000 and 10 000 *R. similis* did not differ significantly from populations recovered on "Yangambi km5" inoculated with 100 *R. similis*. In contrast, the number of *R. similis* recovered from "French Sombre" (*Musa* AAB Plantain sub-group) increased significantly with the inoculum level. The rate of development of *R. similis* on "Yangambi km5" was slower than that on "French Sombre". "Yangambi km5" was also found to be less susceptible to *P. goodeyi* than "French Sombre".

Résumé - *"Yangambi km5" (*Musa *AAA, sous-groupe Ibota) : une source éventuelle de résistance à* Radopholus similis *et* Pratylenchus goodeyi - Des études en pot et en plein champ ont été conduites au Cameroun pour comparer la sensibilité du cultivar "Yangambi km5" à *Radopholus similis* à celle des autres *Musa* triploïdes et évaluer son comportement vis-à-vis de *Pratylenchus goodeyi*. Les résultats des comptages de nématodes au champ effectués tous les 2 mois pendant 2 ans ont montré que les populations de *R. similis* sont significativement plus faibles sur "Yangambi km5" que sur les autres cultivars. L'évaluation en pot de l'incidence de la concentration de l'inoculum sur le degré de résistance du "Yangambi km5" à *R. similis* n'a révélé aucun effet significatif alors que sur le cultivar sensible "French Sombre", nous avons observé des niveaux de populations et de dégâts plus importants avec l'augmentation de l'inoculum. Le développement de *R. similis* sur "Yangambi km5" s'est révélé plus faible que sur "French Sombre". "Yangambi km5" s'est également montré moins sensible que "French Sombre" aux attaques de *P. goodeyi*.

Key words : bananas, genetic improvement, nematodes, plantains.

Bananas and plantains are major staple food crops in parts of Africa, Asia, and South America. Productivity is limited by a number of diseases and pests and nematodes are one of the constraints for which remedies are urgently needed. The major nematode pest species are *Radopholus similis, Pratylenchus goodeyi, P. coffeae, Helicotylenchus multicinctus, Meloidogyne incognita,* and *M. javanica*. In commercial plantations nematicides are used routinely and regularly to maintain productivity (Gowen & Quénéhervé, 1990), but for more than 80% of the world banana production, which includes dessert bananas, cooking bananas, and plantains which are sold and consumed locally, the use of nematicides is not an acceptable control method.

Breeding bananas and plantains for resistance to nematodes is probably the best way of solving the problem for small scale producers. Finding sources of resistance that can be incorporated into acceptable varieties with the necessary resistance to other disease problems remains a difficult long-term objective but should eventually be achievable (Frison *et al.*, 1996). In earlier studies, sources of resistance were found in some diploids of the Pisang Jari Buaya group (*Musa* AA) (Pinochet & Rowe, 1978; Wehunt *et al.*, 1978) and some tetraploids (*Musa* AAAA) bred in Jamaica were not resistant but appeared to have some field tolerance (Gowen, 1976, 1993).

In some preliminary screening conducted in Cameroon, "Yangambi km5", a triploid (*Musa* AAA) cultivar, was found to support the lowest root densities of *R. similis* (Price, 1994).

The objective of this study was to evaluate the response of several cultivars, including "Yangambi km5", to *R. similis* and *P. goodeyi* in pots and under natural infestations in the field.

Materials and methods

Experiment 1: susceptibility of five *Musa* cultivars to *R. similis* in the field

The experiment was conducted at the Centre Régional Bananiers et Plantains (CRBP), Njombé, Cameroon, from May 1993 to January 1995 on land previously cultivated with bananas. Suckers of five cultivars of three genomic groups were taken from the germplasm collection at Njombé. The corms were peeled to remove obvious nematode lesions and root remnants, washed, and then treated with hot water at 55°C for 10-20 min (Blake,1969). Groups of twenty suckers of each cultivar were planted in a randomised block design with three replicates. The cultivars evaluated were:

"French Sombre"	Plantain subgroup *Musa* AAB
Grande Naine	Cavendish subgroup *Musa* AAA
Banane Cochon	Lujugira subgroup *Musa* AAA
Christine	Monthan subgroup *Musa* ABB
"Yangambi km5"	Ibota subgroup *Musa* AAA

Roots were sampled 5 months after planting, then once every 2 months for 16 months. Samples were taken from five plants of each cultivar to form a composite sample. In the laboratory, 25 g subsamples were taken from the washed, chopped and thoroughly mixed composite sample and macerated. The sample was then poured over a column of sieves of 250, 125, 80, and 32 µm and nematodes which collected on the two smallest aperture sieves were counted (Vilardebó & Guérout, 1974).

Experiment 2: effect of *R. similis* inoculum density on responses of *Musa* cultivars

A pot experiment was carried out in a screenhouse at Njombé with cultivar French Sombre (known as susceptible) and the resistant cultivars Yangambi km5 and an Indonesian accession of Pisang Jari Buaya (PJB), a diploid (*Musa* AA). Suckers taken from the germplasm collection were treated as described previously and planted in 10 l containers filled with a sterile mixture of equal parts soil and coffee husks. The soil was of volcanic origin with pH = 5.5. For each cultivar, five suckers or replicates were used for each treatment. Two weeks after planting, *R. similis*, from a population isolated from bananas at Njombé and cultured monoxenically on carrot tissue (O'Bannon & Taylor, 1968), were inoculated at 10^2, 10^3 and 10^4 nematodes per pot. Five plants of each cultivar were included as non inoculated controls. The pots were set out in a completely randomised block design. Watering was done weekly or as necessary. The plants were assessed 6 weeks after inoculation when roots were scored for lesion damage using a technique which expresses area of necrosis as a percentage of root length (Pinochet, 1988; Bridge & Gowen, 1993). Nematodes were extracted as described above.

Experiment 3: lesion formation and *R. similis* reproduction on susceptible and resistant cultivars

In vitro plants of two cultivars, French Sombre and Yangambi km5, recently weaned on a soil coffee husk medium (as above) were planted in 2 l plastic bags. Each plant was inoculated with 10^3 *R. similis* produced from carrot cultures. Plants were sampled 3, 6, 9, and 15 weeks after inoculation; on each occasion, four plants per cultivar were assessed for root necrosis and nematode population densities as described previously. Nematodes were extracted from the total root mass.

Experiment 4: susceptibility of "Yangambi km5" to *P. goodeyi* in the field

Suckers of "Yangambi km5" and "French Sombre" were taken from the germplasm collection at Njombé and treated as described previously. Twenty plants of each cultivar were planted in blocks which were replicated four times. The trial was set up at Bova in south west Cameroon at 850 m above sea level at a site previously cultivated with plantains and known to have an infestation of *P. goodeyi*. Root samples were collected from ten individually marked plants every month for 5 months from the fifth month of planting. Nematodes were extracted as described previously.

Statistical analysis

In all the studies, nematode counts were log-transformed prior to analysis using the Systat 5.2 package; a Duncan's multiple range test was used to separate means when a significant difference was found between treatments.

Results and discussion

Experiment 1: susceptibility of five *Musa* cultivars to *R. similis* in the field

Significant differences ($P \leq 0.05$) in the populations of *R. similis* in different cultivars were found between October 1993 and June 1994 (Fig. 1). In most cases "Yangambi km5" had significantly fewer nematodes than did the other cultivars. Although the plantain "French Sombre" did not differ significantly from the cultivars Grande Naine, Banane Cochon, and Christine, this cultivar consistently showed the highest level of *R. similis* at every sampling. The relative resistance in "Yangambi km5" confirms similar earlier findings (Sarah *et al.*, 1992; Fallas & Marban-Mendoza, 1994; Price, 1994). In contrast with other cultivars, no population increases of *R. similis* were observed over time on "Yangambi km5" and populations never

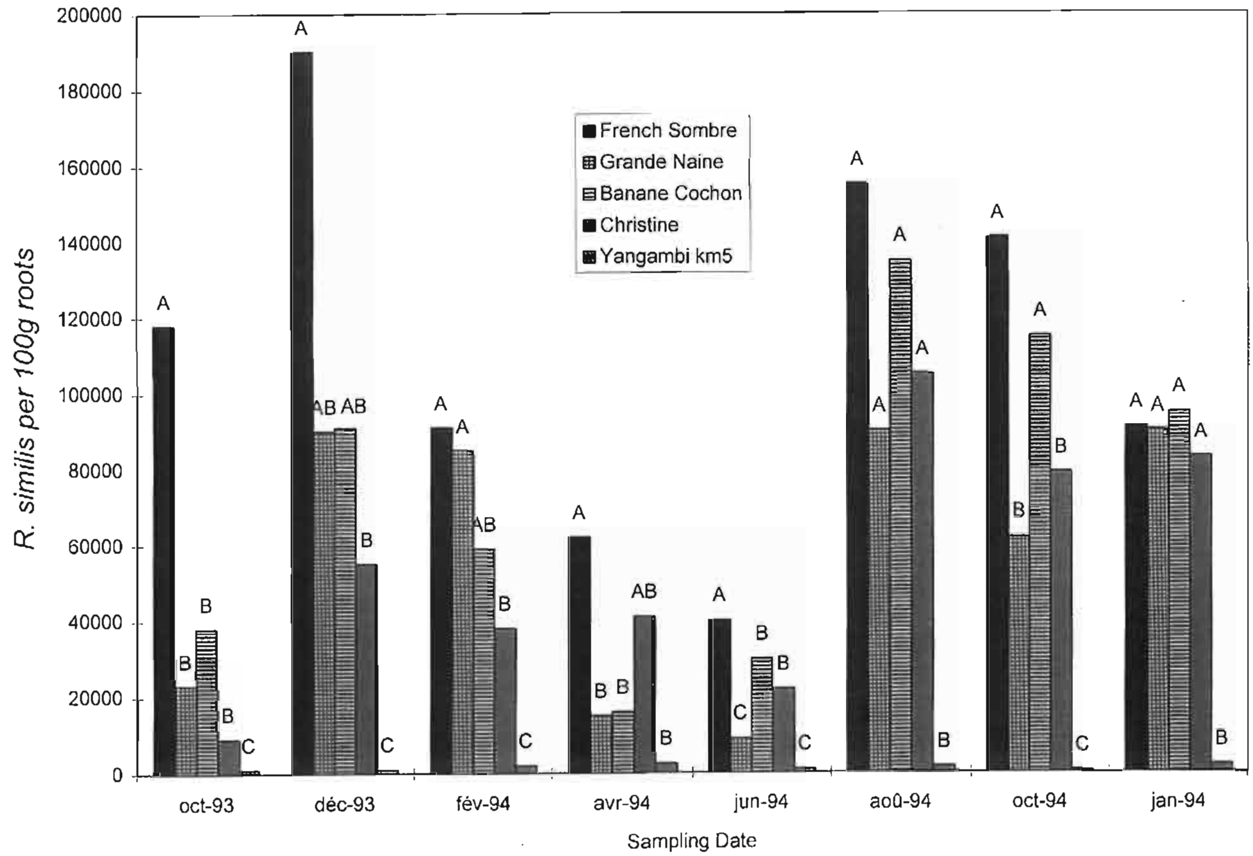

Fig. 1. Comparative population growth of Radopholus similis *in five triploid* Musa *cultivars at different sampling dates in field (Data are means of three replicates; actual data are presented but data were Log [X+1] transformed for analysis; means represented by bars for a sampling date with same letters above them are not significantly different at* $P \leq 0.05$*).*

exceeded 10 000 nematodes per 100 g roots: on susceptible cultivars population densities varied between 10 000 and 150 000 *R. similis* per 100 g roots. The relatively low population increase on "Yangambi km5" indicates that this cultivar is partially resistant to *R. similis*.

EXPERIMENT 2: EFFECT OF *R. SIMILIS* INOCULUM DENSITIES ON RESPONSES OF *MUSA* CULTIVARS

Differences were found between cultivars at the three inoculum levels. "French Sombre" had a significantly higher root lesion index and population of *R. similis* in roots than "Yangambi km5" and Pisang Jari Buaya, which were comparable in their levels of resistance (Table 1). On the susceptible cultivar French Sombre, the population of *R. similis* increased with inoculum level. In contrast, increasing the inoculum concentration had no effect on the level of susceptibility of "Yangambi km5" and Pisang Jari Buaya.

A significant difference was observed between cultivars in the percentages of root lesions 6 weeks after inoculation ($P \leq 0.05$). On "French Sombre", the percentages of necrosis were 26.5, 69.4, and 50.9 for inoculum concentrations 10^2, 10^3, and 10^4, respectively. On resistant cultivars, less than 4% of the total number of primary roots were attacked by *R. similis*, even at the higher inoculum density (Table 1).

EXPERIMENT 3: LESION FORMATION AND *R. SIMILIS* REPRODUCTION ON SUSCEPTIBLE AND RESISTANT CULTIVARS

Nematodes were recovered from roots of both cultivars 3, 6, 9, and 15 weeks after inoculation. No significant difference was observed 3 and 6 weeks after inoculation, suggesting that there was no difference in invasion or penetration of *R. similis*. Pinochet (1988) cautioned that *in vitro* plants should not be used for

Table 1. *Effect of different inoculum levels of* Radopholus similis *on the percentage of infested root, fresh root weight and nematode population build up in roots of three* Musa *cultivars.*

Parameters	Cultivars	Inoculum levels			
		0	100	1000	10 000
Infected roots (%)	"French Sombre"	0	26.6 *a*	69.4 *a*	50.9 *a*
	"Yangambi km5"	0	0 *b*	0.9 *b*	3.4 *b*
	Pisang Jari Buaya	0	0 *b*	0.4 *b*	3.4 *b*
Fresh root weight (g)	"French Sombre"	76 *a*	88 *a*	63 *a*	67 *a*
	"Yangambi km5"	72 *a*	82 *a*	54 *a*	49 *a*
	Pisang Jari Buaya	50 *a*	79 *a*	110 *a*	96 *a*
R. similis/100 g roots	"French Sombre"	0	22 850 *a*	41 100 *a*	83 200 *a*
	"Yangambi km5"	0	0 *b*	0 *b*	450 *b*
	Pisang Jari Buaya	0	100 *b*	300 *b*	900 *b*

Data are means of five replicates. Arithmetic means are presented but data were log transformed for analysis. Means in columns for each parameter followed by the same letter are not significantly different ($P \leq 0.05$).

Table 2. *Percentage of infested root and population build up of* Radopholus similis *on "French Sombre" and "Yangambi km5" 3, 6, 9, and 15 weeks after inoculation with 1000* R. similis *per plant.*

Parameter	Cultivar	Weeks after inoculation			
		3	6	9	15
Infected root (%)	"French Sombre"	22 *a*	30 *a*	50 *a*	76 *a*
	"Yangambi km5"	7 *b*	16 *b*	44 *a*	48 *b*
R,. similis//100 g roots	"French Sombre"	500 *a*	2500 *a*	18 000 *a*	57 000 *a*
	"Yangambi km5"	600 *a*	22 000 *a*	1700 *b*	2600 *b*

Data are means of four replicates. Nematode populations were log transformed before analysis. Means in paired columns followed by the same letter are not significantly different. ($P \leq 0.05$).

resistance studies because of the small size of plantlets and their fragile root systems. Nevertheless, *in vitro* techniques of plant propagation do ensure healthy, uniformly sized plants in which different genotypes can be comparatively assessed. According to Bingfors (1970), Kaplan and Keen (1980), and Dropkin (1989), nematodes normally invade both resistant and susceptible cultivars, and there is often little difference in the numbers of nematodes penetrating root. Nine and 15 weeks after inoculation, the population levels of *R. similis* in roots of "French Sombre" were significantly higher than those from "Yangambi km5" (Table 2) confirming that "Yangambi km5" is a less suitable host for the *R. similis* population used in this study.

A significant difference was observed in the percentages of root necrosis at 3 and 6 weeks after inoculation ($P \leq 0.05$). 20% of the total number of roots of "French Sombre" were attacked, compared to just 7% of those of "Yangambi km5" at 3 weeks after inoculation. On both cultivars, both flecks and large lesions were present, but on "Yangambi km5" 66% of the root system had fleck-like lesions, while on "French Sombre", 66 % of the roots had large tracts of necrosis. Fifteen weeks after inoculation, roots of "French Sombre" had significantly more lesions than "Yangambi km5".

EXPERIMENT 4: SUSCEPTIBILITY OF "YANGAMBI KM5" TO *P. GOODEYI* IN THE FIELD

Pratylenchus goodeyi was recovered from roots of both cultivars at every sampling (Fig. 2). The increase in population was greater on "French Sombre" than on "Yangambi km5" and the populations 8 months after planting were significantly different ($P \leq 0.05$). Ten months after planting, the root populations on "French Sombre" were 45 000 nematodes per 100 g roots, while on "Yangambi km5", the population levels were only 5000.

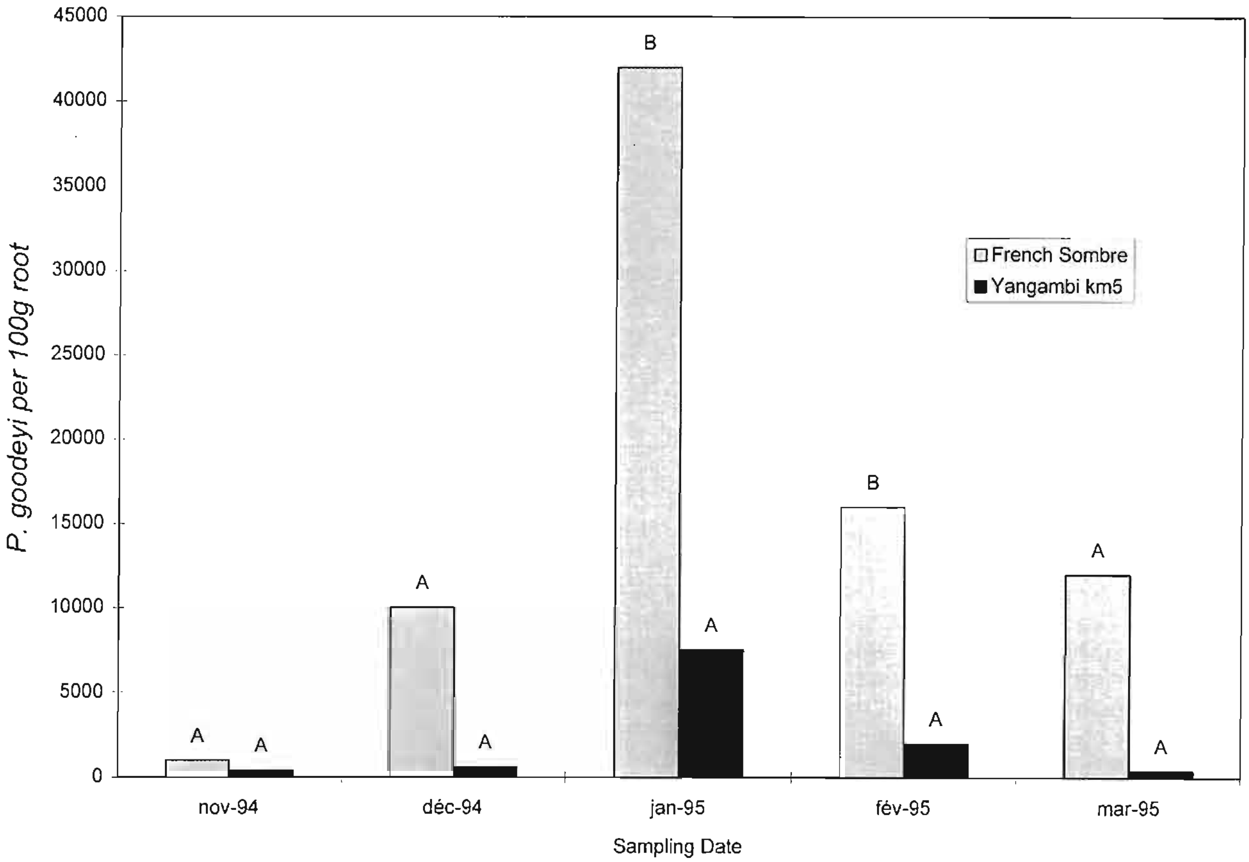

Fig. 2. *Comparative population growth of* Pratylenchus goodeyi *on "Yangambi km5" and "French Sombre" in field conditions (Data are means of four replicates; bars with same letters for the same sampling date are not significantly different at* $P \leq 0.05$*).*

Conclusions

These studies have confirmed that "Yangambi km5" is resistant or partially resistant to *R. similis* and they show that resistance to nematodes does occur in the triploid cultivars. Hitherto, resistance has been found only in diploids which may be more useful in breeding programmes. "Yangambi km5" also has resistance to *P. goodeyi*; it is important that the response to *P. coffeae* is evaluated since the diploid Pisang Jari Buaya is resistant to *R. similis* and not to *P. coffeae* (Pinochet & Rowe, 1978).

Further studies are needed to understand the mechanisms of resistance in "Yangambi km5"; its roots have greater numbers of preformed phenolic cells than Pisang Jari Buaya but the latter has a greater concentration of lignin (Fogain & Gowen, 1996). "Yangambi km5" roots have fewer lacunae than susceptible cultivars (Fogain, unpubl.). Recently, Binks *et al.* (1997) have shown that a Pisang Jari Buaya cultivar produces a phenalenone in response to nematode attack. It appears that resistance to nematodes may involve several mechanisms.

"Yangambi km5" was collected in Zaire; it has other important attributes, particularly resistance to black Sigatoka leaf spot disease, *Mycosphaerella fijiensis* (Fouré *et al.*, 1990; Beverragi *et al.*, 1993), and to the banana weevil *Cosmopolites sordidus* (Fogain & Price, 1994). Another interesting characteristic is its vigorous rooting and suckering particularly in comparison to the plantain subgroup.

"Yangambi km5" has not been found to hybridise readily by conventional breeding and hybridization by seedlings does not look promising (Ortiz, pers. comm.); nevertheless, this cultivar presents various characteristics of such interest that further studies on genetic transfer and on the origin of its *Musa acuminata* components are essential.

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