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Nematode Infection Predisposes Banana to Soil-borne *Xanthomonas campestris* pv *musacearum* Transmission

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ABSTRACT

Xanthomonas wilt is the major disease affecting banana (*Musa* sp.) and enset (*Ensete ventricosum*) production in Ethiopia. A pot trial was carried out to establish the possible role of nematodes in soil-borne transmission of *Xanthomonas campestris* pv *musacearum.* Clean potted 'Pisang Awak' and 'Matooke' plants were first inoculated with a mixed population of nematodes [*Pratylenchus goodeyi*, *Meloidogyne* spp., *Rotylenchus* spp. and *Radopholus similis*] to allow for root damage. After three months, the surrounding soil was drenched with a suspension of *Xcm* (approximately × 10^8 cells per mL) and plants were monitored for Xanthomonas wilt development. A significant disease incidence was observed in plants that had been previously infected with nematodes when compared to those which had not been infected with nematodes. It was concluded that the root damage caused by nematodes creates wounds that act as entry avenues for *Xcm* from the surrounding soil. It was recommended that these results be evaluated on-farm, but in the meantime, an IPM strategy should be adopted for control of Xanthomonas wilt in bananas and enset.

Keywords: banana, nematode, transmission, *Xcm*

INTRODUCTION

Xanthomonas wilt, caused by *Xanthomonas campestris* pv. *musacearum* (*Xcm*) is the main disease threatening banana (*Musa* spp.) and Enset (*Ensete ventricosum*) production in Ethiopia (Addis *et al*. 2004, 2006). Up to 90% yield loss due to Xanthomonas wilt has been reported in 'Pisang Awak' (*Musa* ABB group) plantations in Uganda (Karamura *et al.* 2006). The yield loss incurred on banana and Enset in Ethiopia has not been well documented. Once infected, *Xcm* practically invades all banana parts on a mat that share the vascular system and eventually the banana mat dies (Ssekiwoko *et al.* 2006). Infected Enset plants have also been observed to completely wilt and eventually die. As a result the farmers have abandoned their banana plots in areas with a high incidence of the disease and have resorted to growing other crops such as maize, beans and green leafy vegetables (Bobosha 2005; Shimelash 2006).

Xanthomonas wilt is transmitted in Enset fields through contaminated garden tools used during cultivation (Yirgou and Bradbury 1968; Ashagari 1985; Wolde-Michael *et al.* 2008) and in bananas it is also transmitted by insect vectors via moist male flower cushions and bract scars (Tinzaara *et al.* 2007). In addition, plant-parasitic nematodes are suspected to vector or facilitate the entry of *Xcm* into the plant. Leaf-streak and root-lesion nematodes in particular have been found in association with Xanthomonas wilt diseased plants (Tessera 1989; Swart *et al*. 2000). The nematode species *Helicotylenchus multicinctus*, *Pratylenchus goodeyi*, *Meloidogyne* species and *Radopholus similis* were most frequently found in the banana producing districts of South and South-western Ethiopia (Addis *et al.* 2006). While feeding, the second stage juvenile plant parasitic nematodes pierce root tissues using a stylet (Sarah *et al.* 1996). The resulting wounds are colonised by opportunistic pathogens. These wounds could also act as entry avenues for *Xcm* from surrounding soil since it has been established that plant pathogenic bacteria partly enter their hosts through wounds (Manners 1993). After entry, *Xcm* will spread throughout the whole plant since it is systemic (Ssekiwoko *et al*. 2006). This pot trial was therefore carried out to establish the possible role of plant-parasitic nematodes in the transmission of soil-borne *Xcm* to banana plants through the root system.

MATERIALS AND METHODS

Nematode-infected banana roots (with visible galling and/or root necrosis in the root cortex), were collected from known nematode infested banana plantations (Addis *et al.* 2006) in different localities in South-western Ethiopia. These roots were subsequently cut into 0.5 cm segments which were thoroughly mixed. Nematode inoculation was done by mixing 250 g of nematode-infected root segments with sterilized soil for each pot. Nematodes were extracted from root samples at SARI and sent to NARO, Kawanda, Uganda for species identification. Although nematode population densities of the inoculum were not assessed separately for each pot, a similar plant infection was assumed. The initial nematode population densities per pot were: *Pratylenchus goodeyi* (22,050), *Meloidogyne* spp. (300), *Rotylenchus* spp. (250) and *Radopholus similis* (50). Twenty four (24) pared suckers of each of the *Musa* cultivars 'Pisang Awak' (*Musa* ABB group) and 'Matooke' (*Musa* AAA group) were planted in pots with a diameter and height of 22 cm.

Three months after nematode inoculation, *Xcm*-infected banana pseudostems were chopped into small pieces and these pieces were suspended in a bucket filled with 10 liters of sterilized water. The mixture was stirred intermittently for about one hour to allow the bacterial ooze to discharge into the water. Around the root zone of each plant 300 ml volume of a 2 days old bacterial suspension with a cell concentration of 10^8 cfu/ml (adjusted to $OD_{460\ nm}$ = 0.3 using a spectrophotometer (Bobosha 2005) was poured. The treatments were: (1) only nematode inoculation at

Received: 4 February, 2009. Accepted: 5 April, 2010.

Short Communication

Table 1 Percentage of 'Pisang Awak' and 'Matooke' plants infected with *Xcm* following different levels of exposure to *Xcm* and nematodes.

Treatment	% Plants infected with *Xcm*	
	Pisang Awak	Matooke
Control (neither *Xcm* nor nematodes inoculated)	0 a	0 a
Only nematodes inoculated	0 a	0 a
Only Xcm inoculated	0 a	17 b
Both nematodes and *Xcm* inoculated	50 b	33 c

Means followed by the same letter in a column are not significantly different from each other; according to Tukey's HSD test (P<0.05)

planting, (2) nematode inoculation at planting and *Xcm* inoculation 3 months after planting, (3) only *Xcm* inoculation 3 months after planting and (4) control (no nematode or *Xcm* inoculation). The trial layout was a randomized complete block design with six plants per treatment for each genotype. Hence, a total of 48 plants were assessed in this study. The number of plants developing symptoms characteristic of *Xcm* infection (i.e. yellow wilted leaves and yellow ooze in the leaf petioles or pseudostem) were recorded and were expressed as a percentage of the total plants for that treatment per cultivar. In order to normalize the data, percentage values were Log transformed before separation using Tukey's HSD test (P<0.05) (SPSS 12.0, 2003).

RESULTS AND DISCUSSION

Soil inoculation of *Xcm* around nematode-infected plants led to a significantly higher percentage of disease development compared to soil inoculation of *Xcm* around nematode-free plants (**Table 1**). In the presence of nematodes, 50% of the 'Pisang Awak' and 33% of the 'Matooke' plants developed disease symptoms typical of *Xcm*, while none of the 'Pisang Awak' plants and only 17% of the 'Matooke' plants developed disease symptoms in the nematode-free pots. The plants which were inoculated with only nematodes did not show any visible disease symptoms on the shoot.

Bacteria require either wounds or natural openings such as hydathodes and stomata to enter into a plant (Manners 1993). The presence of nematodes led to damage of roots and the resulting wounds on the root system enhanced entry of *Xcm.* Quimio and Tessera (1996) postulated that *P. goodeyi* could play a role by creating wounds at the time of feeding that could enhance the entry of the *Xcm* bacteria into the plant and could perhaps also play a role in the transmission of the disease. Following nematode damage, it is reported that a number of fungi also invade the root tissue and colonise the stele (i.e. central cylinder) which is usually not damaged by the nematodes (Stover 1972; Gowen *et al*. 2005). This probably hastens the entry of *Xcm* leading to increased disease incidence. An on-farm evaluation of these findings is however recommended, but in the meantime, an IPM strategy should be adopted for the control of Xanthomonas wilt in bananas and enset.

ACKNOWLEDGEMENTS

The authors would like to thank the Flemish Association for Development Cooperation and Technical Assistance (VVOB, Belgium) for its financial support.

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