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Outbreak of basal end rot on banana and plantain in Nigeria

M. MWANGI, R. BANDYOPADHYAY[1], A. TENKOUANO[2] & T. FATUROTI[3]
International Institute of Tropical Agriculture (IITA), P.O Box 7878 Kampala, Uganda
[1]IITA, PMB 5320, Ibadan, Nigeria
[2]IITA, Humid Forest Ecoregional Center, BP 2008 Messa, Yaounde, Cameroon
[3]IITA -Onne, PMB 008 Nchia Eleme, P-Harcort, Nigeria

Abstract In early March 2005 a previously unknown disease was reported on banana and plantain at the International Institute of Tropical Agriculture breeding station located at Onne, S.E. Nigeria. The disease was observed on two varieties, a banana *cv* 25447-S7 and a plantain *cv* 15108-6. Symptoms initiated with dark spots on the peduncle after emergence from the pseudostem. Within two weeks the spots increased in number, expanded in size and spread further down the peduncle towards the fruits. Infection progressed to a dry rot with transverse cracks and shredding of the infected rachis area. Eventually the peduncle broke and the prematurely ripened bunch dropped. Samples of infected plants were taken for laboratory isolation of the pathogen. Fungi recovered included several *Fusarium* species and *Lasiodiplodia theobromae*. Pathogenicity tests indicated *L. theobromae* to be the main causal pathogen of the disease.

Key words: *Musa* spp., peduncle rot, Fusarium species, *Lasiodiplodia theobromae*

Résumé Une maladie inconnue sur la banane a été remarquée début mars 2005 à la station de recherche de l'Institut International d'Agriculture Tropicale située à Onne dans le sud-est du Nigeria. La maladie était observée sur deux variétés, une banane cv 25447-S7 et un type de plantain, cv 15108-6. les symptômes débutaient avec des taches sombres au niveau du pédoncule après l'émergence à partir du pseudostème. Dans une période de deux semaines, les taches augmentaient en nombre, se développaient du point de vue dimension et s'étendait plus à travers le bas du pédoncule, s'étendant vers les fruits. L'infection progressait jusqu'au pourrissement sec avec des craquements transversaux et une déchirure de la région du rachis infecté. Eventuellement, le pédoncule se brisait et le régime tombait, prématurément mur. Des échantillons de plantes infectées étaient récupérés en vue d'une isolation du pathogène en laboratoire. Les champignons isolés comportaient plusieurs espèces de *Fusarium* et *Lasiodiplodia theobromae*. Les tests de pathogenèse indiquaient que *Lasiodiplodia theobromae* était la principale source pathogène de la maladie

Mots clés: *Musa* spp., pourrissement du pédoncule, espèces de *Fusarium*, *Lasiodiplodia theobromae*

Introduction

Banana and plantain constitute one of the most important sources of food and income in the humid forest ecological areas of West and Central Africa (Robinson, 1996). Cultivars grown are mostly triploids and hybrids of *Musa acuminata* and *M. balbisiana*, such as AAB plantains and ABB cooking bananaa. The main producing countries in 2004 included Nigeria, Cameroon, Democratic Republic of Congo and Cote d'Ivore (FAO, 2005).

Most of the production is at low input subsistence level for local consumption, with over 70 million people in West and Central Africa estimated to derive more than one quarter of their food energy requirements from plantains (Robinson, 1996). However, there is increasing production for income generation with enterprises that transform fruits to juices, cakes, flour etc being established in various areas. Constraints to banana/plantain production in the humid forest ecologies include fungal, viral diseases and nematode attacks, poor quality of germplasm, poor crop management and lack of market organization. In an effort to improve productivity efforts continue to be made to breed and disseminate higher yielding and disease resistant varieties (Tenkouano and Swennen, 2004). These efforts are coupled with improvements in integrated management of pests and diseases.

During early 2005 a report of a disease of unknown aetiology attacking plantains and banana was reported within the International Institute of Tropical Agriculture banana and plantain breeding station located at Onne in South East Nigeria. The diseased plants were examined and samples collected for isolation and determination of the cause of the disease. This paper summarises the findings of the diagnostic work.

Materials and methods

The infected fields were visited and data on the population of plants, incidence and severity recorded before sampling. Samples were picked from the peduncle, wilted leaves, pseudostem and corm of the infected plants and taken to the laboratory for isolation of pathogens. From each specimen 20 pieces each measuring 1cm^2 were surface sterilized in 2 % NaOCl for 1 min and rinsed in sterile distilled water. For isolation of fungi ten sterile pieces were plated (5 pieces per plate) on Potato Dextrose Agar (PDA), acidified using lactic acid to pH 4 to reduce bacteria growth. For recovery of bacteria the remaining ten pieces

were plated on Nutrient Agar (NA). The plates were incubated at 24 ± 2 °C. Bacterial growth was assessed after 48 h while fungal growth was evaluated after 5 to 7 days of incubation.

Pathogenicity tests. Six fungi that were isolated from infected plant samples were inoculated onto flowered banana/plantain plants to determine pathogenicity. Pure cultures of the fungi were grown on PDA at 24 ±2 °C for seven days to produce inoculum. The mycelia and conidia formed on PDA were gently scrapped and washed using distilled water to form a suspension, which was used to inoculate plants. A total of 12 banana and 2 plantain plants that had flowered but were uninfected were identified from among plants in the infected fields at Onne. Initially each of the six test organisms was inoculated on two plants, using about 100ml of the fungal spore/mycelial suspension per plant. Two plants were left un-inoculated to act as a control.

Inoculation procedure. On each plant, the exposed peduncle area (between first fingers and point of emergence from pseudostem) was cleaned with water and then washed thoroughly with the inoculum. The inoculated area was covered with paper towels for 4 to 7 days to prevent contamination and washing off of the inoculum by rain. The inoculated plants were examined once per week over a seven-week period, and disease severity estimated using a 0 to 5 scale, indicating between healthy to severe infection, respectively. After seven weeks the inoculated plants were cut down and samples taken from plants showing symptoms for re-isolating pathogens. After obtaining the initial result the pathogenicity test was repeated with two out of the six original fungal isolates selected due to their ability to cause symptoms on inoculated plants. In the repeat pathogenicity tests each of the two fungi was used to inoculate four replicate plants. Disease progress was observed and data recorded as described above.

Results

Symptom description. Initial symptoms were characterised by dark spots on the peduncle between the point of emergence from the pseudostem and the flower. Within two weeks the dark spots increased in number and at the same time coalesced and expanded, spreading further along the peduncle towards the developing fruits. Infected rachis area appeared black and started to shrink followed by a dry rot characterized by transverse cracks. By the third week dark spots appeared on the oldest fruits, leaves nearest to the infected peduncle started wilting and yellowing from the petiole towards the lamina, and eventually collapsed. In advanced stages of disease the infected peduncle area shredded into thin transverse fibers, the fruits started ripening prematurely and developed canker like lesions on the surface.

At the terminal stage the shredded peduncle broke, the bunch dropped to the ground and dieback set in. During periods of continued moist weather sporulation of fungal growth turning from white to orange was observed on the infected peduncle area and on the oldest infected fruits. After sectioning, the infected pseudostem appeared rotted with dark vascular streaking and sometimes emitted a pungent smell. No ooze typical of bacteria infections was apparent in any of the infected plant parts. The vascular discolouration extended downwards to the base of the pseudostem and into the corm.

Disease incidence. The disease attacked only two varieties, one a banana *cv* 25447-S7 and the other a plantain *cv* 15108-6 (Table 1). The two varieties are related in that female parents of both have the same parents. Cultivar 15108-6 was crossed in 1991 and has been grown at Onne since 1993, while *cv* 25447-S7 was crossed in 1995 and has been grown at Onne since 1997. None of the two varieties had shown symptoms of similar infection before this report. Other than at Onne both varieties were being grown at another location (in Ibadan) but there had not been similar infection observed there.

Pathogen isolation. The fungal isolates recovered from the infected plant samples were separated into seven groups based on growth characteristics, spore formation and pigmentation. Four of the seven groups were identified as belonging to different *Fusarium* species while one group of isolates was identified to be *Lasiodiplodia theobromae* (Alexopoulos and Mims, 1979; Agrios, 1988). Two groups that did not sporulate on PDA were not identified. One *Fusarium* isolate was most frequently recovered from different plant parts of both varieties at between 20 to 100% frequency (Table 2). This isolate, identified to be *Fusarium verticilloides*, was recovered from tissues of the infected peduncle area, the pseudostem and also from the prematurely ripened fruits of both infected cultivars. In plants with advanced symptoms

Table 1. Incidence of basal end rot on banana cv. 25447 and plantain cv. 15108 at IITA Onne station.

Field	Variety	Flowered plants[2]	% no. of infected plants
Block PB3	15108-6	9	88.8
Block PB3	25447-S7	22	31.8
OC4 (50m)[1]	15108-6	1	100
OPB (100m)	15108-6	5	0
NC 4 (500m)	15108-6	1	0
Pollinators(20 –30m)	25447-S7	17	5.9

[1]Figure in brackets is distance of field from field PB3 where plants were most infected.
[2]Only flowered plants were considered because symptoms were only visible on the flower part.

Table 2. Isolation frequencies of six fungi recovered from naturally infected banana and plantain at Onne.

Plant part	Cultivar	Isolation frequency (%)[1]						
		Fus[2] A	Fus B	Fus C	Fus F	Lt[3]	Unk[4] E	Unkn G
Peduncle surface[5]	15108	30	100	-	10	-	-	-
Peduncle inner[6]	15108	10	20	-	-	-	-	-
Peduncle surface	25447	-	40	-	-	-	-	-
Peduncle inner	25447	-	20	20	-	-	-	-
Pseudostem	25447	-	60	-	-	-	-	-
Pseudostem	15108	-	-	-	-	10	-	-
Fruits	25447	50	40	40	-	-	100	-
Fruits	15108	-	40	60	10	10	100	10

[1]Frequency is based on total of 10 pieces plated on Potato Dextrose Agar. [2]Fus = *Fusarium* isolates A, B, C and F; [3]Lt = *Lasiodiplodia theobromae*
[4]Unkn = Unidentified isolates E and G which did not sporulate on PDA. [5,6]Samples taken from surface or at 2 cm depth on the infected peduncle area.

Lasiodiplodia theobromae was recovered at 10% frequency from the infected pseudostem and ripening fruits of cv 15108-6. However, after inoculation *Lasiodiplodia* was re-isolated at higher frequencies of up to 30 % on rachis, pseudostem and the infected fruits but only when symptoms were not very advanced (data not shown).

Pathogenicity tests. Disease symptoms were only observed on the plants inoculated with *Fusarium* isolate B or with *Lasiodiplodia theobromae*. With each of these fungi infection had set in three weeks after inoculation, but disease was more severe on plants treated with *L. theobromae* than on those inoculated with *Fusarium*. By the 7th week after inoculation infection was at terminal stages with bunch fallen on plants treated with *L. theobromae* while on plants treated with *Fusarium* B symptoms were not as severe. From the samples of the inoculated plants, five groups of fungi were re-isolated with *Fusarium* isolate B again the most frequently isolated followed by *L. theobromae*.

Discussion

Pre-harvest diseases of the banana fruit caused by fungi are important because they produce blemishes that are unacceptable to consumers. Although some of these infections may not affect the eating quality, they lower marketability of fruits. The disease that is reported here was different in that it affected the fruit in the pulp making it inedible. A peduncle rot of bananas has previously been reported (Jones and Stover, 2000), but the symptom progression and infection pathway was different from those of the disease reported here. In the reported cases which were in Australia and Central America, the infection started from the end of peduncle where the male bud was broken off and moved upwards towards the fruits. Infection that we diagnosed started on the region of the peduncle before the oldest fingers and moved downwards towards the fruits and also backwards into the pseudostem and through it to the corm.

Several fungi and bacterial isolates were recovered from infected plant tissues. Most of the fungi were identified to belong to *Fusarium* species and one group was identified as *Lasiodiplodia theobromae* (Alexopoulos and Mims, 1979). Based on morphology and growth habit the most frequently isolated *Fusarium* isolate **B** was identified as *F. verticilloides*. Although *Fusarium* was most frequently isolated, it was *Lasiodiplodia* that caused the most severe symptoms and killed plants fastest.

In Australia several *Fusarium* species and *Verticillium theobromae* have been reported to be associated with infected peduncles (Muirhead and Jones, 2000). In Central America and in the Phillipines *L. theobromae* was the pathogen mostly isolated from infected samples (Jones and Stover, 2000). It is likely that a majority of the organisms recovered from infected plant samples are saprophytes that rapidly colonize infected tissues. In plants with advanced symptoms it was difficult to isolate *Lasiodiplodia*, because it was often overrun by the faster-growing *Fusarium* species. However, after inoculating on plants *Lasiodiplodia* was re-isolated successfully from tissues that were not too heavily diseased. It was concluded that *L. theobromae* was the causal pathogen of this disease, which as far as we know, has not been previously reported in Africa.

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