Field Diagnostics and Surveillance Manual

for banana Fusarium wilt



TABLE OF CONTENTS

Overview	1
What is banana Fusarium wilt?	
What is field diagnostics?	
What is disease surveillance?	
What is needed for field diagnostics?	
What is needed for disease surveillance?	
Banana Fusarium wilt	2
What causes banana Fusarium wilt?	
How does it spread?	
How can it be identified?	
Symptoms of banana Fusarium wilt	
Collection of samples	7
Collection of samples and GIS information	
Collecting field information on Fusarium wilt	
Sanitation and prevention of spread	10
Isolation of Foc from collected material	11
Isolating the fungus from discoloured vascular strands	
Single-sporing of isolates	
Maintenance of healthy cultures	
Shipment of cultures	
Steps involved in laboratory diagnosis of Fusarium wilt of banana	13
Methods to prepare culture media for Foc	15
Half-strength potato dextrose agar	
Carnation leaf agar	
Methods for maintaining Foc cultures	15
Storage on sterile filter paper	
Storage on CLA slants	
Deep-freezing	
Storage in soil	
Lyophilisation of Fusarium cultures	
Steps for Foc TR4 surveillance in Mozambique	17
How can Fusarium wilt be controlled?	
Things you can do to prevent spread of Fusarium wilt	
Valuable additional reading	19

Overview

What is banana Fusarium wilt?

Banana Fusarium wilt (or Panama disease) is the most destructive disease of bananas in the world. The disease is caused by a soil-borne fungus which infects banana roots and blocks the movement of water and nutrients in the plant. Eventually the plant wilts and dies. Banana Fusarium wilt was first discovered in Australia, but gained prominence when it destroyed thousands of hectares of Gros Michel in Central America during the early 1900s. The disease was brought under control when Gros Michel was replaced by Cavendish varieties. For the past 25 years, however, Cavendish bananas have been severely affected by a new Asian strain of the fungus. This strain, called Foc TR4, has been discovered in Mozambique in 2013. Foc does not affect crops other than banana, and is not toxic to humans.

What is field diagnostics?

Identification of a disease based on symptoms and signs, and by considering environmental factors and production practices. Field diagnosis has to be confirmed by laboratory work.

What is disease surveillance?

Disease surveillance is an exercise by which the spread of a disease is monitored in order to establish patterns of progression. The main role of disease surveillance is to predict, observe, and minimise the harm caused by the outbreak, as well as to increase knowledge about which factors contribute to such circumstances. A key part of modern disease surveillance is the practice of disease case reporting. Early detection, spread prevention and destruction of infected plants are the only effective means to contain Fusarium wilt caused by Foc TR4.

What is needed for field diagnostics?

• Knowledge of symptoms and signs of banana Fusarium wilt

What is needed for disease surveillance?

- Machetes, knifes, scissors
- Paper bags and envelopes
- Gloves and disinfectants
- Paper/notebooks
- Vehicles, GPS and cameras

Banana Fusarium wilt

What causes banana Fusarium wilt?

Fusarium wilt is caused by *Fusarium oxysporum* f. sp. *cubense* (Foc), a fungus that can survive for decades in infested soils. Foc is classified into three races according to the variety of bananas that they affect. These include Foc race 1 (infects Gros Michel, Pisang Awak and Apple banans), Foc race 2 (infects cooking bananas such as Bluggoe) and Foc race 4 (infects most banana varieties, including Cavendish bananas). Foc race 4 is further divided into tropical (TR4) and subtropical (STR4) strains, of which Foc TR4 is the most severe.

Foc also consists of vegetative compatibility groups (VCGs). By knowing the VCG of a specific strain of Foc, it is possible to determine its origin and spread. Foc TR4 includes VCG 01213/16, which originated in Southeast Asia. The VCG has now been introduced into several other countries outside Asia, including Mozambique, Oman, Pakistan, Lebanon and Jordan.

VCG	Race	Origins
0120/01215	1, 4	Australia, Brazil, Costa Rica, France (Guadeloupe, Guiana), Honduras, Indonesia (Java), Jamaica, China, Malaysia (Sarawak), Nigeria, Portugal (Madeira), South Africa, Spain (Canary Islands), Taiwan, USA (Florida)
0121	4	Indonesia (Sumatra, Kota), Taiwan
0122	4?	Philippines
0123	1	Malaysia (peninsular and Sarawak), Philippines, Taiwan, Thailand, China
0124/	1, 2	Australia, Brazil, Burundi, China, Cuba,
0125/		Democratic Republic of Congo, Haiti,
0128/		Honduras, India, Jamaica, Malawi,
01220/		Malaysia, Mexico, Nicaragua, Rwanda,
01222		Tanzania, Thailand, Uganda, USA (Florida), Zanzibar, Kenia
0126	1?	Honduras, Indonesia (Irian Jaya, Sulawesi), Papua New Guinea, Philippines, China
0129/01211	1, 2	Australia
01210	1	Cayman Islands, Cuba, USA (Florida)
01212	?	Tanzania
01213/01216	4	Australia, Indonesia (Agam, Dharmasraya, Halmahera, Irian Jaya, Java, Solok Sulawesi, Sumatra, Pariaman, Tanag Datar), Malaysia (peninsular),Taiwan, Jordan, Oman, Mozambique, China, Philippines
01214	None	Malawi
01217	None	Malaysia
01218	None	China, Indonesia (Java, Sumatra), Malaysia (peninsular), Thailand
01219	None	Indonesia (Java, Sumatra)
01221	None	Thailand, China
01223	None	Malaysia
01224	None	

Table 1: The vegetative compatibility among strains of Fusarium oxysporum f. sp. cubense

Sources: Jones (2000); Ploetz (2005b); Lodwig et al. (1999).

How does it spread?

Foc can spread with infected planting material and in soil and water. It can also be transmitted from the parent plant to the suckers, or between plants via root contact. Anything that moves soil, such as machinery and equipment, vehicle, tools, clothing and footwear, can carry and spread the fungus.

How can it be identified?

- Symptoms and signs of the disease in the field
- Morphological identification of spores and hyphae produced by Foc
- Pathogenicity testing by inoculating healthy banana plants with the fungus
- Molecular identification by using of DNA markers
- Phenotypic identification by VCG analysis

Symptoms of banana Fusarium wilt

The first sign of Fusarium wilt is the irregular yellowing on the edges of older leaves of affected plants (Fig. 1A). The yellowing then progresses to the youngest leaves. Leaves eventually turn brown and collapse to hang down the pseudostem. Splits can often be observed in the lower pseudostem of affected plants (Fig 1C). Once all the leaves have died and collapsed, the plant is unable to fill bunches. Suckers from the mother plant, however, can still be healthy (Fig. 1B).

Since other abnormalities might also result in the yellowing of banana leaves, it is often necessary to investigate internal symptoms. When cut down, yellow to reddish-brown streaks can be found in the outer layers of the pseudostem (Fig. 1E). Further dissection of the plant shows a prominent discolouration of the inner rhizome, which could either affect a small section or the entire inner rhizome, depending on the age and severity of the infection (Fig. 1D).

Fusarium wilt should not be identified on external symptoms alone, as this could lead to misidentifications. Different banana varieties express symptoms differently (Fig. 2). For instance, yellowing is often not visible on Gros Michel bananas affected by Foc race 1, while symptoms caused by Foc race 1 on Pisang Awak leads to bright yellow leaves. Bacterial wilt can also cause yellowing of banana leaves, but usually on the youngest leaves. Erwinia and the banana weevil, however, can cause yellowing of older banana leaves. Yellowing can also be caused by abiotic factors such as water-logging, nutrient deficiencies and even lightning (Fig. 3). It is, therefore, important to properly investigate banana plants turning yellow and by taking a sample to confirm the cause of the yellowing as Fusarium wilt. Also, banana diseases should also be inspected for important diseases present in east and central Africa, such as banana Xanthomonas wilt and the banana bunchy top virus (Fig. 4).

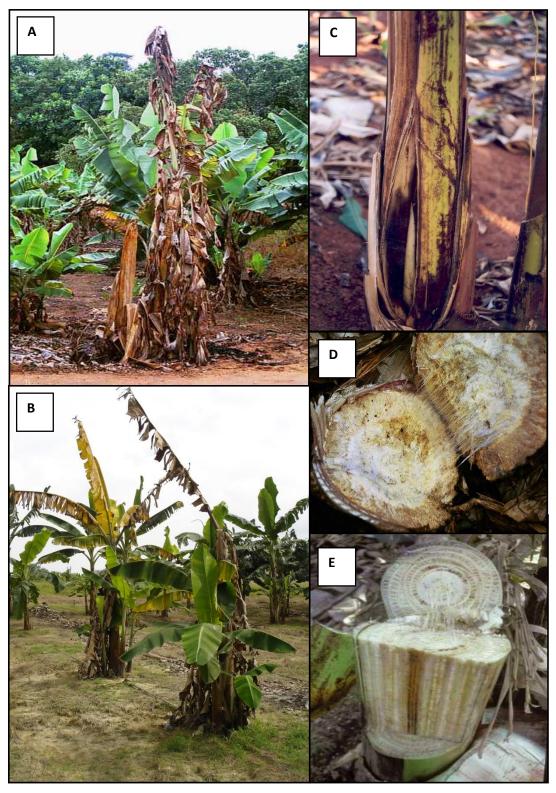


Figure 1. Disease symptoms of Fusarium wilt of banana caused by 'subtropical' race 4 of *Fusarium oxysporum* f. sp. *cubense* (Foc) in South Africa (A) and 'tropical' race 4 in Malaysia (B). Affected plants wilt rapidly, older and then younger leaves become yellow and brown, and plants eventually die. In some cases, the base of pseudostems split (C). Internally, a deep golden discolouration of the inner rhizome develops (D), while the vascular bundles in the pseudostem will turn yellow to reddish-brown (E).

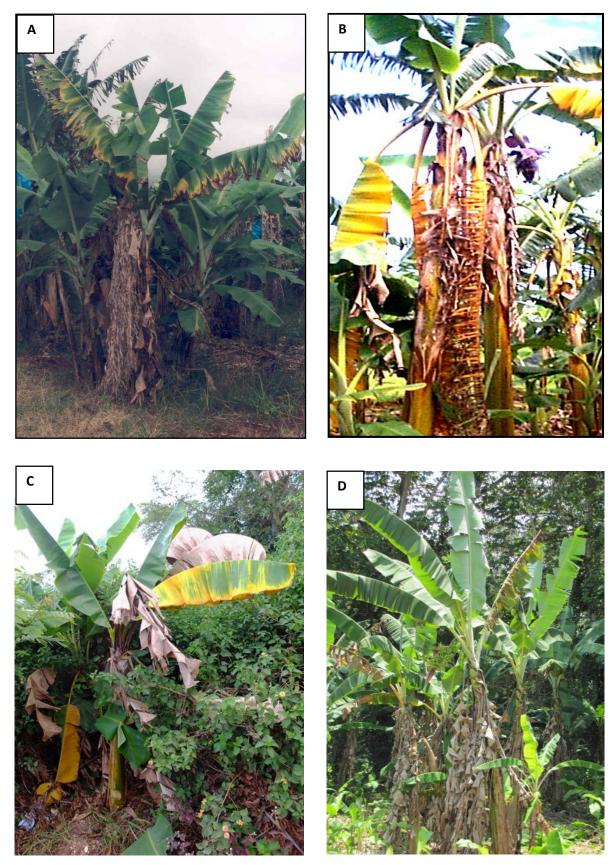


Figure 2. Disease symptoms of Fusarium wilt on different banana varieties: (A) Cavendish bananas in South Africa, (B) Pisang Awak in Uganda, (C) Pisang Awak in Mozambique, and (D) Gros Michel in Costa Rica.

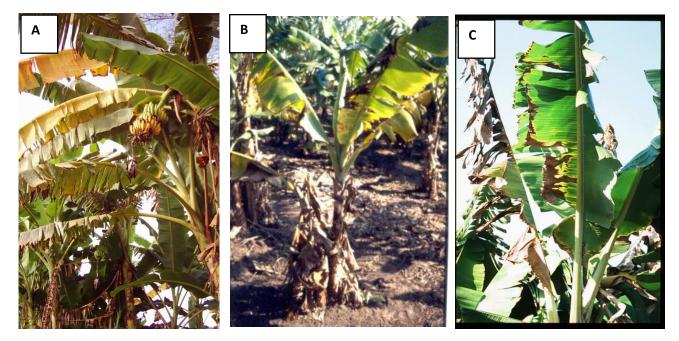


Figure 3. Yellowing of banana leaves caused by (A) Banana Xanthomonas wilt (bacteria), (B) the banana weevil borer, and (C) nutritional deficiencies.

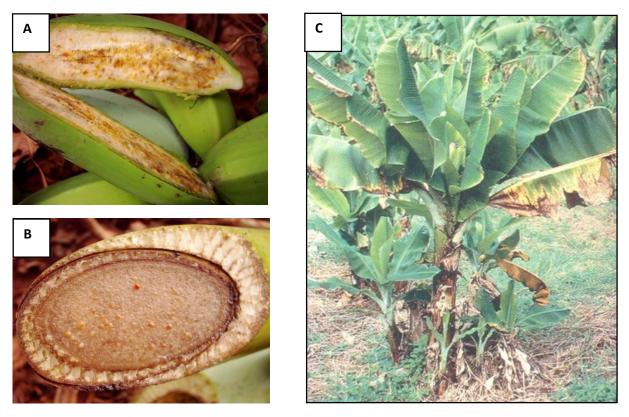


Figure 4. External symptoms caused by *Xanthomonas campestris* pv. *musasearum* (Fig. 3A) is reminiscent of banana Fusarium wilt, but with younger instead of older leaves turning yellow. Internal discolouration of fruit (A) and oozing of bacteria from stems and stalks (B) clearly distinguish the disease from Fusarium wilt. Banana bunchy top disease (C) is recognised by the very characteristic stunting of plants.

Collection of samples (Courtesy N. Moore)

The sample should consist of a section from the pseudostem of the wilted banana plant where typical continuous discoloured vascular strands are evident (Fig 5). The sample should be taken from as low in the pseudostem as is possible but not from areas where decay is advanced. Also, the sample should be taken from as close to the centre of the pseudostem as is possible, as opposed to the outermost leaf bases. The chance of recovering healthy Foc cultures decreases as the sample deteriorates. Samples should be wrapped in paper towel before and after the strands are excised. Avoid plastic bags as this promotes growth of bacteria.

Ideally, the discoloured vascular strands should be dissected at the time of sampling or as soon as possible thereafter. The use of sterile blotting papers is recommended and aseptic technique should be applied to the dissection of strands. Where several samples are being prepared, a fresh piece of blotting paper should be used for each sample, and scalpel blades should be flamed if possible or at least wiped with 70% alcohol between samples. The excised strands should then be placed between sterile blotting papers in a paper envelope to dry naturally. Do not let the strands get too hot (e.g. in direct sunlight or in the boot of a car) as this may kill the fungus. Fusarium wilt specimens do not need to be kept in the fridge, and should be kept dry.

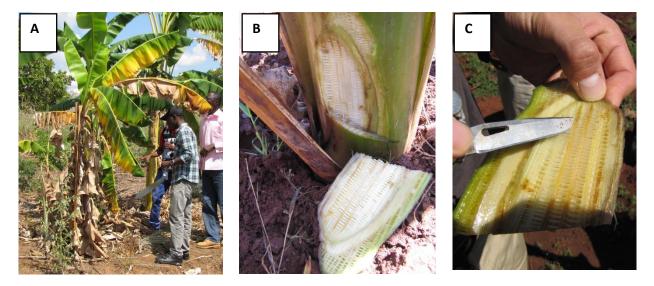


Figure 5. Vascular strands need to be collected from pseudostems of banana plants with typical Fusarium wilt symptoms (A). This can be achieved in a non-destructive way (B) by slicing open part of the pseudostem on the side of yellow leaves, and by dissecting out discoloured xylem tissue (C).

In large commercial plantations, owners of banana plants are advised to contain Fusarium wilt by killing the diseased and surrounding plants, and by fencing the plants that were killed. In northern Mozambique, as in many African countries, small growers cannot afford to kill and fence in affected plants. It is thus recommended that the importance of Fusairum wilt be explained to the owner and the community, and that the diseased tissue be shown to them. Diseased plants should be killed, burned and/or buried (Fig. 6).



Figure 6. Interaction with the owner and community in handling banana Fusairum wilt is important. After sick plants are discovered they should be killed, burnt and/or buried.

Collection of samples (information to be added on envelope):

Accurate notes must be taken for each sample including:

- Sample number (one sample number per plant)
- Date
- The variety of the host plant, including local names (and uses if known)
- Genomic constitution of host if known (e.g. AA, AAB, ABB etc.)
- Age of plant/plantation
- Whether plants sampled are grown in a garden, commercial plantation, village or the wild.
- Size of the diseased area where the plant was collected, with photos
- Location (e.g. name of province/state, how far in what direction from nearest town, name of road, name of property if sample is from a commercial plantation etc.)
- Collectors names
- Other useful observations might include the source of the planting material, whether the plant is growing in water-logged soil, how many plants are affected, what other varieties are growing in the vicinity and are these diseased or healthy?

Collecting field information on Fusarium wilt

Table 2. Information should be collected at all sampling sites, irrespective of the presence of the disease (see example below)

Date	Sampling route	Sample number	Banana variety	Status +/-	Identity	Collector	GPS coordinates
19 Aug	Namialo	0001	Pisang Awak	+			

Table 3. To investigate dissemination of Fusarium wilt, owners/producers need to provide

 information on the history of the plantation and production practices to surveillance officers

Information available and actions taken	Yes	Partly	No	Don't know	Comment
Knowledge of cropping history, including diseases					
Regularly inspecting plants for pests and diseases					
Would be able to identify symptoms in fact sheet					
Awareness of quarantine regulations					
Financial resources to deal with disease outbreaks					
Use of tissue culture bananas					
Use of bits and suckers from other farms					
Use of fertilisers from credible source					
Machinery and equipment is shared with others					
Tools and equipment on-farm is cleaned regularly					
Visitors to the farm require permission					
Traders can enter premises without restrictions					
Animals move through plantation					
Land is affected by floods					
Irrigate plantation from rivers and streams					
Other information					
Whether plants sampled are grown in a garden, commercial plantation, village or the wild.					

Size of the diseased area where the plant was collected, with photos.

Sanitation and prevention of spread

Risks for spread of Foc TR4 by inspectors during surveillance will be minimised as follows:

- Clean boots and/or protective wear over shoes will be used during sampling (Fig. 7)
- Each team will ensure that vehicles are properly cleaned and free of soil after visiting banana farms and informal plantings (Fig. 8)
- Shoes, knives and all other equipment used during sampling will be comprehensively cleaned and washed between the different banana plantations/plantings surveyed
- No vehicles of inspectors will be used in plantations
- The two Foc TR4-affected farms will be visited last



Figure 7. To prevent wet soil and mud sticking to shoes, plastic bags can be placed over shoes



Figure 8. Both vehicles and shoes shoed be disinfected between visits of banana farms

Isolation of Foc from collected material (Courtesy N. Moore)

Isolating the fungus from discoloured vascular strands

Isolation can be attempted when the strands have dried. Small sections (3-6 mm long) of dry discoloured vascular strands are submerged into plates of ¼ strength potato dextrose agar (PDA) medium amended with an antibacterial agent (e.g. streptomycin @ 1.2 mL/240 mL PDA). If present, Fusarium growth will appear from the strands in 2 to 4 days. However, if the sample is badly contaminated with bacteria this may mask fungal growth. Let samples dry further if this occurs and increase the strength of the antibacterial amendment in the media. A high rate of recovery of Fusarium should be expected from correctly prepared samples. Single-spore (monoconidial) cultures should be prepared from an isolate from each specimen.

Single-sporing of isolates

Single-spore isolates of *F. oxysporum* are obtained by either dilution plating or streaking (demonstrated below). For both methods, a small scrape of sporulating hyphae are collected from cultures grown on ¼-strength PDA plates, and dissolved in 10 ml sterile distilled water in test tubes. From the initial spore suspension, a series of dilutions can be prepared. One ml of each of the dilution series is then either pipetted or streaked onto water agar, and the water agar plates incubated with the lid up overnight at 25°C. The plates are viewed for germination of conidia under a dissecting microscope the following morning, and single-conidia cut from the water agar with a surface-sterilised scalpel and transferred to new 90-mm ¼-strength PDA plates. Additionally, single-spore cultures can also be obtained by dissecting the very tip of single growing hyphae from an older culture grown on CLA.

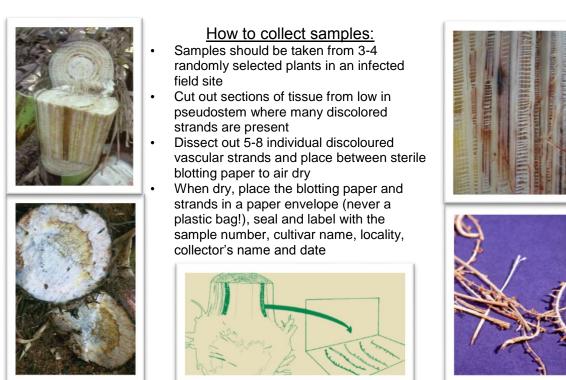
Maintenance of healthy cultures

Healthy (sporodochial-type) cultures of Foc from single spores are maintained on carnation leaf agar (CLA) to prevent mutation. Cultures can be initiated on weak-strength PDA medium (e.g. ¼ strength) to check the morphology of cultures for taxonomic purposes or for spore production. Healthy cultures of Foc growing on PDA medium exhibit abundant fluffy aerial mycelium after 2 days and produce abundant microconidia. Some macroconidia may also be produced on PDA although this type of spore is more commonly produced on CLA medium. Cultures of Foc should NOT be kept on PDA medium for longer than 4 or 5 days as mutations can rapidly occur and these cannot be reversed. Mutated cultures (e.g. slimy pionnotal mutants) should be discarded. Cultures are normally maintained in an incubator at 25°C. Black light is generally not required for cultures of Foc to sporulate. Various methods are used for long-term (e.g. lyophilisation), medium-term (e.g. colonised filter paper in cold storage) and short-term (e.g. CLA) storage of cultures of Foc.

Shipment of cultures

If posting the strands for isolation and analysis, please post in a paper envelope as soon as the strands are dry enough with sample numbers and details clearly written on or with each sample envelope. Please include a copy of the relevant quarantine import permit inside the package if this is required.

Note: If there is any possibility that samples have been mixed up and the details for some samples may be incorrect, discard the samples concerned.



Preparation of wilt samples

Figure 9. To send cultures for identification, they should be prepared according to standard methodology, and sent to the receiver in paper envelopes clearly marked with all the details of the collection site.

Steps involved in laboratory diagnosis of Fusarium wilt of banana

- Receive specimen, log details and observations from grower/inspector.
- Isolate from symptomatic tissue (usually 2 x Strep' PDA with 4 pieces per plate).
- Check morphology of resultant growth (macro and microscopically) (Fig. 10).
- Subculture Fusarium and make spore suspension for single-sporing onto water agar.
- Select 2 x germinated single spores to initiate monoconidial cultures.
- Assign accession number to isolate and record in *Fusarium* isolate database.
- The monoconidial culture can be maintained as follows (Fig. 11):
 - a) Filter paper culture for medium term storage
 - b) CLA plates or slants for short-term storage
 - c) Lyophilisation for long-term storage in collection
 - d) Glycerol (15%) for long term storage
 - e) Soil samples for long term storage

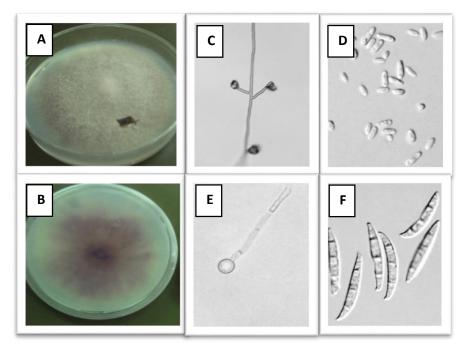


Figure 10. Cultural and morphological features of Fusarium oxysporum f.sp. cubense:

(A and B) Top and bottom side of a culture grown on PDA, (C and D) Microconidia borne in false heads and in water, (E) chlamospores, and (F) macroconidia.

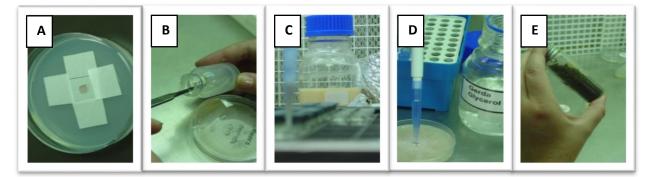
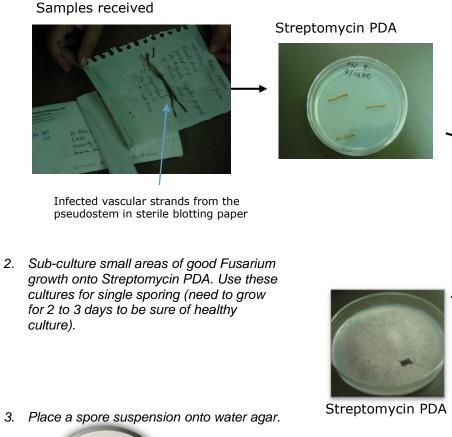


Figure 11. Storage of *Fusarium oxysporum* f. sp. *cubense*: (A) Filter paper, (B) CLA slants, (C) Lyophilisation, (D), 15% Glycerol and (E) In soil.

1. Isolation from plant material.



3. Place a spore suspension onto water agar.

Small cube of If isolation plates are not heavily contaminated, a shortcut can be taken by using Fusarium growth from the isolation plate for the spore suspension. 4. After 24 hours take at least 2 single germinated spores and place back onto Streptomycin-PDA. Streptomycin-PDA Small cube of agar with a single germinated spore

5. Once single spore cultures exhibit normal Fusarium growth (should be visible after 4 to 5 days), chose only 1 single spore culture to represent each isolate and discard all other cultures. Assign unique accession number. Subculture onto CLA, which becomes the source for VCG, DNA and pathogenicity tests and filter paper culture for storage, and for extra CLA cultures if required for freeze drying/lyophilisation.

Methods to prepare culture media for Foc

Half-strength potato dextrose agar

Mix 19 g of Potato Dextrose Agar (PDA) and 10 g of agar with 1000 mL of distilled water in a 1-L Schott bottle. Sterilise in an autoclave (wet cycle: 100 kPa at 121°C for 20 min). When cooled to 50 °C, add 1.2 mL streptomycin solution (1g streptomycin sulfate powder per 100 mL sterile distilled water) and shake well. Pour the PDA into 90-mm-diameter Petri dishes and allow to solidify. When set, the plates are stored upside down in a refrigerator at 4°C.

Carnation Leaf Agar

Mix 20 mL agar with 1000 mL of distilled water, autoclave and pour into Petri dishes as described above. Add one to two pieces of sterilised carnation leaves onto the surface of freshly poured water agar plates after the medium has set. Store as described above.

Methods for maintaining Foc cultures

Storage on sterile filter paper

Filter papers disks (5 cm in diameter) are autoclaved in glass petri dishes. The disks are then aseptically placed on ¼-strenth PDA in petri dishes. *Fusarium* isolates are then cultured on CLA for 7-10 days. Rectangular agar pieces colonised by *F. oxysporum* (3 mm in diameter) are then placed on the sterile filter papers, and grown for 7-10 days, until the entire filter paper is covered by mycelia. The filter paper with fungal growth is lifted off the PDA, placed in sterile petri dishes, and left for 1 day to dry. It is then cut into smaller pieces (5 mm in diameter) and placed in cryovials. The cryovials are all clearly labelled with the isolate number, and stored at 5°C until use.

Recommended period of storage: 3 months - 1 year

Storage on CLA slants

Water agar (WA) is prepared by dissolving 20 g agar in 1 L distilled water. The WA is then autoclaved at 121°C for 20 minutes. After autoclaving, aliquots of 10 ml of the WA are poured into sterile 20-ml bottles under sterile airflow. The bottles are placed in a tray, and the tray kept at an angle of 45°C until the WA is solidified. A single sterile carnation leaf is placed on top of the agar. The isolate is then placed next to the carnation leaf on the water agar, and grown at 25°C for 1 week. All cultures are clearly marked with the isolate number, and stored at 5°C until further use.

Recommended period of storage: 3 months - 2 years

Deep-freezing

A 15% glycerol stock solution is first prepared and autoclaved. The *F. oxysporum* isolates are then grown on $\frac{1}{2}$ -strength PDA plates at 25°C for 7-10 days. Ten ml of the 15% glycerol are then pipetted onto the fungal growth in the petri dishes in a sterile flow cabinet. The spores and some hyphae are carefully dislodged with a surface-sterilised scalpel. One-ml-aliquots of the spore suspension are then pipetted from the petri dishes into 2-ml cryovials. Each of the cryovials are carefully labelled, placed into cryovial boxes and stored at -80°C. When the isolate needs to be recovered, small quantities of the frozen suspension is scraped from the cryovial with a sterile scalpel, and placed onto the culture medium.

Recommended period of storage: Up to 5 years

Storage in soil

Soil is first sterilised in small glass bottles or tubes. The cultures are then grown on ½-strength PDA plates for 7-10 days. Sterile distilled water (20 ml) is poured onto each culture in a flow cabinet, and the spores discretely dislodged with a surface-sterilised scalpel. Ten ml of the fungal spore suspension is then pipetted from the petri dishes, and aseptically transferred onto the soil in the glass bottles and the tubes. All the glass tubes and bottles are clearly marked with the isolate number, and stored at room temperature. The isolate is recovered by placing a small amount of soil onto culture medium.

Recommended period of storage: Up to 5 years

Lyophilisation of *Fusarium* cultures

Isolates to be lyophilised are grown on carnation-leaf agar in petri dishes for 7-10 days. Several colonised carnation-leaf pieces are then transferred to each of five replicate sterile 5-ml glass vials labelled with the isolate number. A 0.5-ml aliquot of sterile skim milk is added to each vial. The vials are then stoppered with split rubber stoppers, which allow for evacuation of air. The stoppered vials are placed in a tray and quickly frozen by pouring liquid nitrogen into the tray. A Lucite plate slightly larger than the tray is placed on top of the partially stoppered vials.

A drying chamber on a refrigerated freeze-dryer is used for lyophilisation. The tray is placed on the pre-cooled shelf in the drying chamber, and a vacuum is pulled. When refrigeration is completed, the heat is turned on, while the samples dry gradually. After lyophylisation, the vials are sealed under vacuum by inflation of the rubber diaphragm in the chambers over the tray, which presses down the Lucite plate and forces the rubber stoppers to seal the vials. After lyophylisation, vials are capped and labelled, and the vials stored at -20° C.

Recommended period of storage: Up to 20 years

Steps for Foc TR4 surveillance in Mozambique

- 1. Before inspection of properties:
 - Obtain permission from the Ministry of Agriculture and owners to survey their properties
 - Provide the background and process of the surveillance to property owners
 - Obtain information on the history and production practices of bananas
 - It is an offence to obstruct an inspector or any other authorised person to conduct surveillance on properties planted to bananas
- 2. Finding suspect plants:
 - Look for plants with yellow or wilted leaves and/or stem splitting
 - Sampling must be conducted on all suspect plants
 - Suspect plants should be marked with flagging tape or spray paint. The end of the row or block should also be marked to find the suspect plant again
 - Plants should only be cut down if superficial internal symptoms in the pseudostem cannot be observed in the suspect plants
 - Felling plants could prevent further investigations if laboratory tests are inconclusive
- 3. Reporting suspect plants:
 - Banana growers must report plants suspected of being affected by Foc TR4
 - Any links to risk items (e.g. plant material, vehicles, machinery) must be reported
- 4. Sampling and testing:
 - Plants with external *Fusarium* symptoms must be checked internally. This may require cutting down the pseudostem, even into the rhizome.
 - Several samples should be taken from the same plant for sending to the laboratory
 - Suspect plants should be clearly marked and isolated
- 5. Reporting of results:
 - Test results need to be provided to the property owner before making them known
 - If Foc TR4 is identified, the Mozambique Department of Agriculture will be informed to further deal with the outbreak
- 6. What happens when results are positive:
 - The property will be placed under quarantine by the Department of Agriculture
 - Quarantine signs will be placed at the entrances to the property, and the movement of people without the owner's permission will be discouraged

- Infected properties may be permitted to continue growing, harvesting and selling bananas but under strict conditions to prevent spread of the disease
- The Department of Agriculture will regularly inspect the property to ensure that all requirements for containment of Foc TR4 are met
- 7. Decontamination of surveillance teams:
 - All efforts to minimise risk should be taken by the surveillance teams. This includes comprehensive cleaning and decontamination exercises to prevent spread.

How can Fusarium wilt be controlled?

Since the discovery of Fusarium wilt of banana, various control methods have been attempted to curb the damage caused by the disease. Yet, no long-term control measures are available other than the use of resistant banana varieties. Soil fumigation, the use of fungicides, biocontrol products and sterilants, crop rotation, flood-fallowing and organic amendments are some of the control strategies that have been investigated in the past, without much success. Current management practices include the use of disease-free tissue culture plantlets, avoiding sharing farm equipment with other growers, preventing the introduction of the disease in disease-free areas, and detecting and isolating outbreaks early.

There are things you can do to prevent spread of Panama disease

- Know if you are in a Panama disease infested area
- Know how to recognise Panama disease
- Report all new outbreaks
- Prevent the movement of people and vehicles by isolating infected areas.
- Do not transport infected or contaminated material to areas that are free of the disease
- Keeps the spread of Panama disease out of unaffected areas at all cost.

Valuable additional reading

- Beckman, C.H. 1987. The nature of wilt disease of plants. The American Phytopathology Society, St. Paul, MN.
- Booth, C. 1971. The Genus Fusarium. Commonwealth Mycological Institute, Kew, Surrey, England.
- Nelson, P.E., Toussoun, T.A. and Marasas, W.F.O. 1983. *Fusarium* species: An illustrated manual for identification. The Pennsylvania State University Press, London.
- Ploetz, R.C. 1990. Fusarium wilt of bananas. ASP Press, St. Paul, Minnesota, USA.
- Nelson, P.E., Toussoun, T.A. and Cook, R.J. 1982. *Fusarium*: Disease, Biology and Taxonomy. Pennsylvania State University Press.
- Snyder, W.C. and Hansen, H.N. 1940. The species concept in *Fusarium*. American Journal of Botany 27: 64-67.
- Stover, R.H. 1962. Fusarial wilt (Panama disease) of bananas and other *Musa* species. Commonwealth Mycological Institute, Kew, Surrey, UK.