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Field survey guidance for *Fusarium oxysporum* f. sp. *cubense* Tropical Race 4

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The IPPC Secretariat and the IPPC community are grateful to all the institutions that contributed to the production of this document. This document presents guidance to national plant protection organizations (NPPOs) to support active surveillance, detection, diagnostics, control and prevention of *Fusarium oxysporum* f. sp. *cubense* Tropical Race 4 (TR4). It was created with technical and financial support from developed countries partnering with the IPPC, in the framework of the Africa Phytosanitary Programme (APP), and reviewed by technical officers at the IPPC Secretariat. The IPPC implements APP in collaboration with the Food and Agriculture Organization of the United Nations (FAO) and the African Union Commission on Agriculture, through its technical unit, the African Union Inter-African Phytosanitary Council (AU-IAPSC).

Abbreviations

APHIS	Animal and Plant Health Inspection Service
APP	Africa Phytosanitary Programme
AU-IAPSC	African Union Inter-African Phytosanitary Council
FAO	Food and Agriculture Organization of the United Nations
FOC TR4	<i>Fusarium oxysporum</i> f. sp. cubense Tropical Race 4
GIS	Geographic Information System
IPPC	International Plant Protection Convention
NPPO	National Plant Protection Organization
SR4	Subtropical Race 4
USDA	United States Department of Agriculture

Introduction

Consistent and thorough monitoring and surveillance are essential for countries to determine the presence or absence of plant pests, particularly those that significantly affect food security, the environment, trade, and agricultural productivity. This early warning information is vital for prompt pest outbreak response, making sound phytosanitary decisions, effective risk management, and safeguarding borders against pest entry and spread.

This field survey guidance therefore provides easy-to-follow guidelines for technical personnel of national plant protection organizations (NPPOs), to survey *Fusarium oxysporum* f. sp. *cubense* Tropical Race 4 (FOC TR4). FOC TR4 is a major pest threatening global banana production, can cause 100 percent yield loss on a farm and is challenging and costly to manage.

This document provides a protocol, with visuals, to aid in the monitoring, detection, sample collection, and diagnostics of FOC TR4, ensuring effective phytosanitary decision-making to manage the pest risk and protect trade in plants and plant products.

The field survey guidance complements the digital tools available to NPPO plant health inspectors, through the Africa Phytosanitary Programme (APP) mobile application and Geographic Information System (GIS) platforms.

APP is an initiative of the International Plant Protection Convention (IPPC) designed to transform pest management across Africa by enhancing the capabilities of phytosanitary personnel within NPPOs, to leverage advanced science and modern digital technology for effective and timely pest surveillance, detection, diagnostics, control, and prevention.

The IPPC implements APP in collaboration with the Food and Agriculture Organization of the United Nations (FAO) and the African Union Department of Agriculture, Rural Development, Blue Economy and Sustainable Development, through the African Union Inter-Africa Phytosanitary Council (AU-IAPSC).

Figure 1. Adult banana plant infected with FOC TR4 exhibiting leaf yellowing and wilt



© Scot Nelson / USDA

Field survey guidance for *Fusarium oxysporum* f. sp. *cubense* Tropical Race 4

Scientific name

Fusarium oxysporum f. sp. *cubense*
Tropical Race 4 (E.F. Sm.)
W.C. Snyder & H.N. Hansen

Common name

FOC TR4, Fusarium wilt of banana,
Panama disease

Type of pest

Fungus

Taxonomic position

Phylum: Ascomycota
Class: Sordariomycetes
Order: Hypocreales
Family: Nectriaceae

Known hosts

Preferred hosts

Gros Michel banana (*Musa acuminata*),
M. acuminata subsp. *burmannica* and
M. balbisiana. Tropical Race 4 (E.F. Sm.)
W.C. Snyder & H.N. Hansen

Survey protocol

Survey-site selection

Survey where *Musa* species occur.

This may include commercial production sites, landscaped areas or natural areas with wild *Musa* plants.

Time of year to survey

Year-round; however, older leaves are more likely to have symptoms of the disease. Banana suckers that are less than about four months old (often used as planting material) do not develop visible symptoms.

Visual survey

Signs and symptoms

The first above-ground symptoms are pale green streaks at the bottom of the leaf petioles at the base of the oldest leaves. After this, two types of syndromes can occur:

YELLOW-LEAF SYNDROME

- ◆ The leaves begin to yellow, progressing from older (bottom) to younger (top) leaves, and may turn brown at the edges (see Figure 2).
- ◆ The leaves gradually collapse, bend and droop, forming a skirt around the base of the plant (see Figure 4).
- ◆ This process can last one to three weeks.

GREEN-LEAF SYNDROME

- ◆ The leaves remain mostly green until they bend and collapse; older leaves exhibit this syndrome before younger ones.

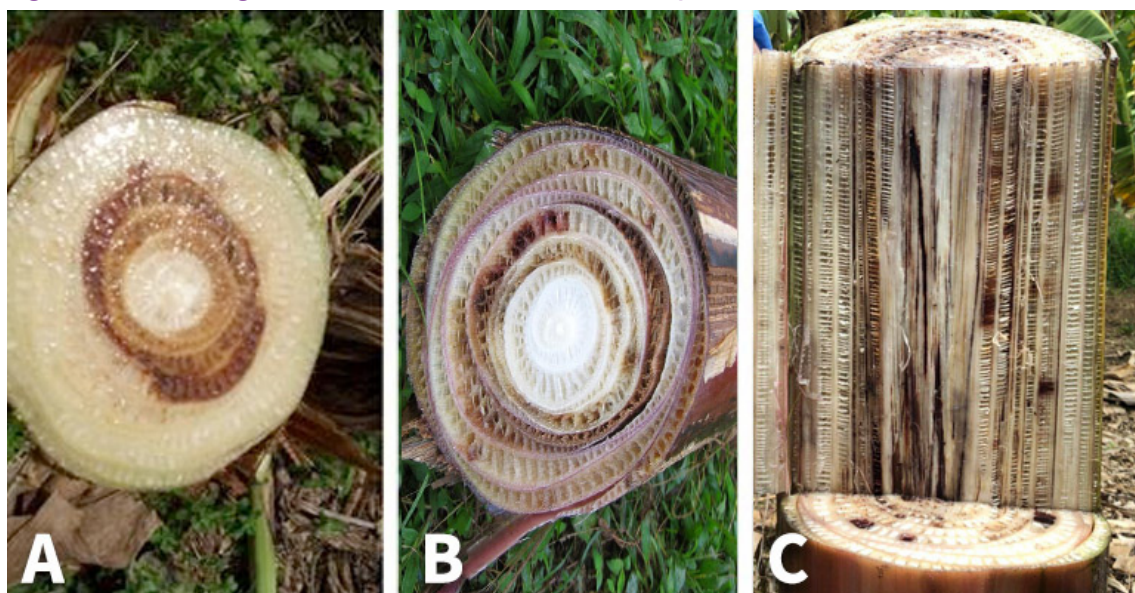
The pseudostems (the part of the banana tree that looks like a trunk) may split lengthwise and, when cut open, a reddish-brown discolouration of the vascular tissue is typically seen (see Figure 3). A reddish-brown discolouration may also be seen in the roots and rhizomes. New leaves may be paler and shorter than normal. Fruits exhibit no symptoms.

Figure 2. Early stage of FOC TR4 infection – leaf yellowing and edges turn brown



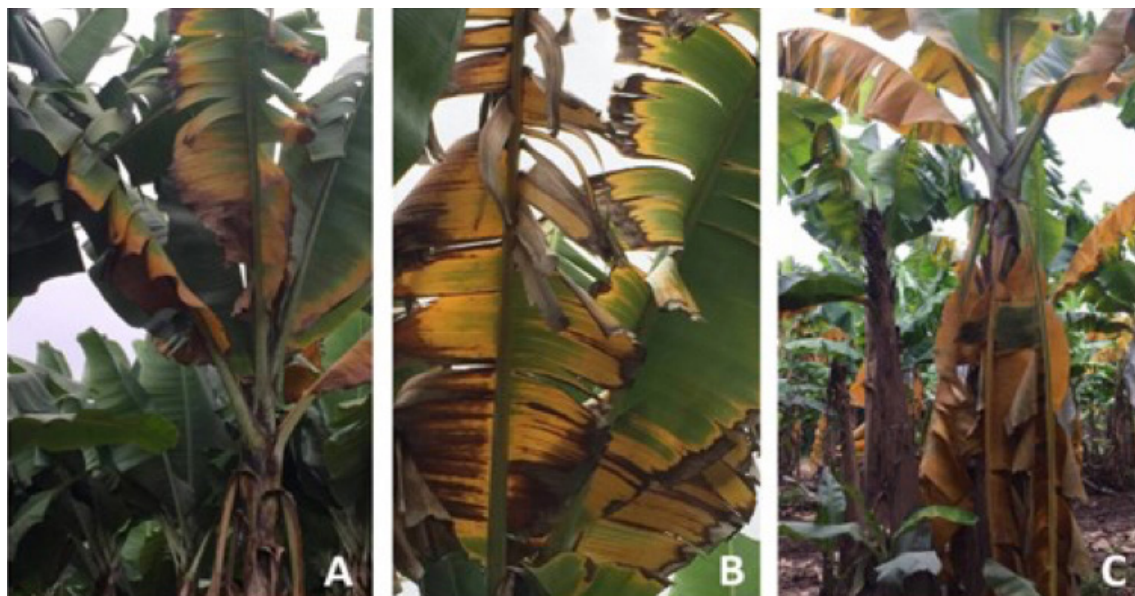
© Department of Agriculture and Fisheries, Queensland, Australia

Figure 3. Various stages of internal discoloration in banana pseudostems as a result of FOC TR4



3(a) © George Mahuku/ IITA; 3(b) © Department of Agriculture and Fisheries/ Queensland, Australia 3(c) © Miguel Dita /Alliance of Bioversity and CIAT

Figure 4. Advanced stage of FOC TR4 infection: (a) leaves wilt and start to collapse; (b) advanced yellowing and browning of leaves; (c) dead leaves form a skirt around the stem



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Associated organism

The banana weevil, *Cosmopolites sordidus* (see Figure 5), is a suspected vector of this pathogen. The presence of this insect could be an indication that the pathogen may be present.

Sample collection

Disposable gloves are recommended for sampling. If sampling multiple plants, gloves and cutting tools should be sanitized (or the gloves changed) between plants to avoid cross-contamination. Sample the pseudostem of the plant where continuous discoloured vascular areas (strands) are present. Sample as low as possible on the pseudostem (closer to the ground) but not from areas where decay is advanced. Avoid sending only the outermost leaf bases of the pseudostem. A small (5 cm × 5 cm) piece of rhizome tissue showing discoloured vascular strands can also be used, but only if decay is not advanced. Banana tissues are very wet and can degrade quickly in hot weather. Plastic bags are not recommended as they may quicken the deterioration of the sample. Place the sample in a clean, dry paper towel or newspaper in a heavy paper bag. Make sure the sample is accurately labelled for the follow-up. Keep the sample cool and dry in an ice chest, but do not freeze. If sampling between fields where FOC TR4 is suspected, remove the soil from footwear or use disposable foot covers between fields to avoid spreading the pathogen.

Pictures of symptoms from the field may also help identifiers to determine the location, the overall plant health and how the samples looked before they were removed from the host.

Pest identification and diagnostics

Pest description

Fusarium oxysporum f. sp. *cubense* (FOC) has four races, three of which affect banana. Race 4 has two subgroups: TR4 and subtropical race 4 (SR4), which infect hosts in the tropics and subtropics respectively. Cold-stressed banana plants grown in subtropical regions are predisposed to SR4 outbreaks, whereas TR4 is an aggressive strain that can infect banana plants that are healthy or unstressed.

F. oxysporum f. sp. *cubense* TR4 is a soil-borne pathogen that is found within the vascular tissues of host plants. The pathogen can survive in plant residue and can persist in soil for up to 40 years. The fungus is commonly spread through infected planting material, infested soil and water, or possibly through the banana weevil. Infected banana plants can also transmit the pathogen via underground rhizome growth to new banana plants. The total process, from the initial external symptoms on a plant to collapse and death, can take three to six weeks.

F. oxysporum f. sp. *cubense* TR4 is like other FOC races and produces three types of asexual conidia that can only be seen with a microscope. Microconidia (5 µm to 7 µm × 2.5 µm to 3 µm) are oval or kidney-shaped spores that can be produced under any conditions and are the most produced spore within infected plants. Macroconidia (22 µm to 36 µm × 4 µm to 5 µm) are larger, thin-walled spores, most frequently produced on the surface of infected plants. The third group, chlamydospores (9 µm × 7 µm) are round, thick-walled spores that are produced in macroconidia or are intercalary or terminal in older hyphae.

Figure 5. Adult banana weevil, *Cosmopolites sordidus* (Germar)



© G. McCormack, Cook Islands Biodiversity Database

Identification and diagnostic resources

The different races of FOC cannot be distinguished by symptoms alone and therefore laboratory techniques are needed for identification. The recommended molecular method to screen for FOC TR4 is polymerase chain reaction (PCR) using the W2987-F and W2987-R primers that amplify DNA specific to FOC TR4.

Mistaken identities

Besides the other races of *Fusarium oxysporum* f. sp. *cubense* that affect banana, the fungus causes symptoms that may also be confused with other bacterial diseases of banana such as banana xanthomonas wilt (*Xanthomonas campestris* pv. *musacearum*), moko disease of banana (*Ralstonia solanacearum* race 2) and blood disease of banana (*Ralstonia syzigii* subsp. *celebesensis*). However, these pathogens cause symptoms on fruit (which FOC does not) and can have noticeable bacterial ooze from freshly cut pseudostems (which FOC would not). FOC TR4 symptoms may also be confused with other biotic and abiotic factors, including water stress. Care should be taken to examine the plant for other external and internal symptoms.

IPPC

The International Plant Protection Convention (IPPC) is an international plant-health agreement that aims to protect global plant resources and facilitate safe trade. The IPPC vision is that all countries have the capacity to implement harmonized measures to prevent pest introductions and spread, and minimize the impacts of pests on food security, trade, economic growth, and the environment.

Organization

- » There are over 180 IPPC contracting parties.
- » Each contracting party has a national plant protection organization (NPPO) and an official IPPC contact point.
- » Ten regional plant protection organizations have been established to coordinate NPPOs in various regions of the world.
- » The IPPC Secretariat liaises with relevant international organizations to help build regional and national capacities.
- » The secretariat is provided by the Food and Agriculture Organization of the United Nations (FAO).

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