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Tropical race 4 of Panama disease in the Middle East

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Background Panama disease (aka Fusarium wilt) of banana (*Musa* spp.) has been a destructive problem for well over a century. Race 1 of the pathogen, *Fusarium oxysporum* f. sp. *cubense* (Foc), was responsible for the demise of the first export trades of banana that were based on the cultivar 'Gros Michel'. Currently, tropical race 4 (TR4) impacts the Cavendish cultivars, which are most important in both export and smallholder production. TR4 was confirmed in Jordan in 2013, but has probably been present in the country since at least 2005. The outbreak in Jordan was apparently the first occurrence of Panama disease in the Middle East, but it also represented a considerable expansion of TR4's distribution, which had previously been restricted to the Far East. How TR4 arrived in Jordan is not known. However, it is clear that TR4 has spread within Jordan, and is now also present elsewhere in the Middle East and Africa. We review the history, epidemiology and management of Panama disease, and discuss the current distribution of TR4 and its potential impact on banana production in the Middle East.

Keywords Fusarium wilt of banana · Panama disease · *Fusarium oxysporum* f. sp. *cubense* · *Musa* spp · Cavendish

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Introduction

Banana (*Musa* spp.) is arguably the world's most important fruit crop. In 2011, combined global production was about 145 million tons with a gross production value of US$44 billion (FAOSTAT 2013). About 15% of all production reaches international markets, virtually all of which are from cultivars in the Cavendish subgroup. The remaining 85% is sold in local markets, and a large portion of these fruit (ca 28% of the total) are also from Cavendish cultivars. Thus, one of the 50 recognized subgroups of banana accounts for over 40% of all production (Ploetz & Evans 2015).

Despite banana's reputation as a crop that is grown in the humid tropics, it is important and has had a long history in the arid, subtropical Middle East. By the 12th century, banana was present throughout northern Africa and in Moorish Spain, preceding the crop's introduction to the Eastern New World in the 1500s. Arab traders probably played a key role in its dissemination in the Middle East, and the name "Musa" may have originated from the Arabic term for banana, "mauz" (DeLanghe 2002). In 2012, about 3 million tons of fruit were harvested in the Middle East (Table 1). Production was recorded in 16 countries or territories, and with the exceptions of Pakistan and Jordan, production increased in each of the major producers (i.e. producers of more than 35,000 tons) between 2002 and 2012.

In general, the edible bananas are hybrids between and among *M. balbisiana* and different subspecies of *M. acuminata* (Perrier *et al.* 2011). The bananas that were first cultivated in the Middle East were probably hybrids between these species, since *M. balbisiana* is more tolerant of drought and cold temperatures than *M. acuminata* and interspecific hybrids would, thus, be better adapted to conditions in the region (DeLanghe 2002). Nonetheless, pure *M. acuminata* cultivars, notably the Cavendish clones, were introduced to the region during the last century (Brown 1908; DeLanghe 2002). Due to their productivity and despite their more demanding requirements, Cavendish cultivars are now widely grown in the region; most of what appears in Table 1 comes from this closely related set of genotypes.

Table 1 Production of banana in the Middle East in 2012

Country	Tons of fruit harvested	% change since 2002
Egypt	1,129,777	+28.7
Sudan (former)	750,000	+76.9
Morocco	222,267	+36.9
Turkey	206,346	+117.2
Pakistan	135,000	- 5.5
Israel	129,522	+34.4
Yemen	127,468	+31.3
Lebanon	125,000	+84.6
Oman	63,000	+91.4
Jordan	38,852	- 18.0
Cyprus	6,901	- 34.3
Palestinian Authority	4,000	- 26.5
Bahrain	1,000	+17.2
Algeria	335	n/a
Syria	249	- 66.0
United Arab Emirates	200	+52.7
Total	2,939,917	

Data from FAOSTAT, 2013.

Panama Disease

Panama disease (aka Fusarium wilt) is one of the most destructive diseases of banana (Stover 1962; Stover & Simmonds 1987). Its common name recognizes the extensive damage it caused in export plantations in this Central American country beginning in 1890 (Stover 1962). By 1960, Panama disease had spread widely in tropical America, the Caribbean and Western Africa, destroying 40,000 hectares of 'Gros Michel' (Ploetz 2005; Stover 1962). These epidemics played a significant role in the trades' abandonment of 'Gros Michel' and their conversion to the Cavendish subgroup. Currently, the latter cultivars are the most important bananas in both export and smallholder production worldwide.

Panama disease is caused by *Fusarium oxysporum* f. sp. *cubense* (Foc), which is a member of the *F. oxysporum* species complex (Fosc) (Michielse & Rep 2009; O'Donnell *et al.* 2009). The Fosc contains nonpathogens, as well as plant and animal pathogens (di Pietro *et al.* 2003; Michielse & Rep 2009). Plant pathogens in the Fosc often exhibit considerable host specificity, and single pathogenic forms, the formae speciales, affect a single or limited set of host plants.

Four pathotypes (aka "races") of Foc are recognized on banana: race 1, which caused the epidemics on 'Gros Michel'; race 2, which affects ABB cooking bananas, such as 'Bluggoe'; subtropical race 4 (SR4) that affects Cavendish and race 1 and 2 suscepts in the subtropics; and TR4, which affects many of the same cultivars as SR4, but in the absence of disease-predisposing cold temperatures that occur in the subtropics (Ploetz & Pegg 2000). Race 3 of Foc does not affect banana. Although better understandings of variation in Foc are needed, it appears that TR4 is comprised of a single clone of the pathogen, vegetative compatibility group (VCG) 01213-01216 (Ploetz *et al.* 2015). Dita *et al.* (2010) were able to develop a diagnostic procedure for TR4 that relied on the homogeneous nature of the pathogen and it has been used to diagnose new outbreaks of this destructive race (Garcia *et al.* 2014).

Distribution and epidemiology

Panama disease is thought to have originated in Southeast Asia, where Foc coevolved with its banana host (Ploetz & Pegg 1997; Stover 1962; Vakili 1965).

Distinct, clonal lineages that are recognized in Foc reflect its polyphyletic composition (Fourie *et al.* 2011; Koenig *et al.* 1997; O'Donnell *et al.* 1998). Although an uncommon lineage of the pathogen was found exclusively in Malawi (lineage C5 in O'Donnell *et al.* 1998 and V in Koenig *et al.* 1997), an overwhelming majority of strains of this pathogen worldwide can be assigned to lineages that are present in Southeast Asia (Fourie *et al.* 2011; Koenig *et al.* 1997; O'Donnell *et al.* 1998). Simmonds' (1966) suggestions that "wherever a susceptible clone has been grown in bulk, the disease has appeared" and that "the susceptible clone can generate its own pathogen" have not been corroborated.

The pathogen is disseminated in infected seedpieces (rhizomes, aka "corms" or "suckers"), soil, surface waters that are used for irrigation, contaminated tools, farm equipment, clothes and footwear (Stover 1962). Tissue culture plantlets are free of Foc, but can be infected if grown later in infested soil, either in the field or in nurseries.

Once Foc is established in a given soil, it can survive for lengthy periods in the absence of its banana host. Reports of 20 years are common (Stover 1962), and in extreme cases 40 years have been indicated (Simmonds 1966; Buddenhagen 2009). Stover (1962) suggested that resilient chlamydospores of Foc were responsible for its longevity, but its survival as a non-pathogenic parasite of weed roots (Hennessy *et al*. 2005; Waite & Dunlap 1953) is probably more important for long-term survival in the absence of its banana host (Ploetz 2015).

Due to its diverse means of dispersal and long-term survival in infested soil, Foc may be the most widely disseminated of all banana pathogens. In a relatively recent summary, Stover & Simmonds (1987) indicated that Panama disease was present in all banana-producing regions except islands in the South Pacific and Melanesia (i.e., the Bismarck and Solomon Islands), Somalia, and countries surrounding the Mediterranean Sea.

Although VCG 01213-01216 was first identified in samples from Taiwan in 1990, the presence of a "new" Cavendish-destroying race was only recognized as monocultures of these cultivars began to succumb in Indonesia and Malaysia (Ploetz 2006b). In many cases, the origins, original distribution, and the timing of and means by which TR4 spread in Southeast Asia were unclear (Ploetz *et al.* 2015). Although Foc-contaminated surface waters that were used for irrigation were evidently responsible for the rapid development of epidemics in China, Malaysia, and the Philippines, it is not known how and why Cavendish plantations were affected in Sulawesi and other areas in the region that were supposedly free of the pathogen and were planted with tissue culture plantlets (I.W. Buddenhagen, personal communication).

By the turn of the century, TR4 was recognized in a fairly wide area that included Australia (Northern Territory), China (Hainan, Guangdong and Guangxi), Indonesia (Halmahera, Irian Jaya, Java, Sulawesi and Sumatra), Malaysia (Peninsular and Sarawak), the Philippines (Mindanao) and Taiwan (Ploetz 2006a; b). In 2013, TR4 was confirmed for the first time outside Southeast Asia (Butler 2013; Garcia *et al*. 2014). The first outbreaks in Jordan and Mozambique were soon followed by confirmations in Lebanon, Oman and Pakistan (Fusarium wilt of banana 2015; Ordonez *et al*. 2015).

Clearly, TR4 is capable of long-distance dissemination. However, the means by which this has occurred are currently unclear. Given the size of banana suckers and the great distances that would have been involved, it seems unlikely that infested/infected plants were responsible for its movement from Southeast Asia to Africa and Western Asia. Although something as simple as muddy boots of plantation workers from Southeast Asia may have been responsible, more information is needed on how this pathogen could survive such an arduous passage. Preventing new outbreaks of TR4 may require better understandings of its long-distance dissemination.

Management

There are limited options for managing Panama disease. The perennial pathosystem and polycyclic nature of the disease have complicated the development of measures that are effective for several years or fruiting cycles (Ploetz, 2007; 2015). Moreover, poor resistance exists in important groups of banana, and technical hurdles confront those who would improve disease-susceptible cultivars. Susceptible banana cultivars can usually be grown only if pathogen-free propagation materials are used in pathogen-free soil. Tissue culture plantlets are the most reliable source of clean material, but they are more susceptible to Panama disease than traditional banana seed pieces (Smith *et al.* 1998). The latter trait may have facilitated the spread of TR4 and development of the epidemic in Jordan (see below). Nonetheless, plantlets should be used to propagate this crop whenever possible.

In pathogen-free regions, effective quarantine measures are most important. Programs have been created in the Western Hemisphere to ensure that stakeholders are informed about the symptoms and potential impact of TR4 (Pocasangre *et al*. 2011), and a regional contingency plan was published by the Organismo Internacional Regional de Sanidad Agropecuaria (OIRSA) (Dita Rodríguez *et al.* 2013), which details recommended responses if TR4 were ever introduced to the Western Hemisphere. Similar plans would be beneficial in the Middle East. Foc cannot be eradicated from infested soil, and after it moves into a region its exclusion from noninfested plantings can be difficult. Early recognition and delineation of new outbreaks are essential.

Biological control of Panama disease has received considerable attention, but long-term efficacy of this approach in the field has not been demonstrated (Ploetz 2015). Diverse microbes have been tested, but most reports are on *in vitro* assays or short-term greenhouse studies. Field studies are uncommon and in no case has acceptable control been reported for this perennial crop. Annual losses of 10-20% that occur with the best biocontrol treatments may be tolerable in a short-term crop (*e.g.* tomato or radish), but are not sustainable in a multi-year banana crop (Ploetz 2015).

Limited or questionable efficacy has been associated with chemical and physical measures that have been tested against this disease (Ploetz 2015). Surface disinfestants have been identified that eliminate the pathogen from infested tools, but effective prophylactic or therapeutic fungicides are not known. Nitrogen forms (NO_3 vs NH_4) and silicon have been studied for disease suppression, but only minor reductions of this disease have been reported after their supplemental use. Rice hull burning (heat sterilization of the soil) has been recommended in the Philippines and Indonesia, but no efficacy data are available. Other soil treatments, such as solarization, methyl bromide fumigation, and flood fallow are usually effective for no more than a single fruiting cycle, since Foc rapidly recolonizes treated areas (Herbert & Marx 1990; Hermanto *et al*. 2012; Stover 1962). Finally, soils that suppress the development of Panama disease are known. However, they are poorly understood, and the responsible factors have not been identified and used to convert disease conducive soils to suppressiveness.

Mixed plantings in which diverse banana cultivars are grown with other crops usually develop only moderate losses compared to monoculture situations (Stover 1962). Thus, when high yields of a single cultivar are not the objective, it may be possible to produce susceptible clones in heterogeneous plantations. In contrast, monoculture production of susceptible cultivars is difficult in infested areas. In Foc-infested soils, resistant cultivars have been the only consistently effective tool for managing this disease.

Resistant cultivars exist for several different kinds of banana, but are needed in other situations (Buddenhagen 1990). There is a critical need for TR4-resistant bananas that meet standards imposed by local and export markets (Ploetz & Evans 2015). Xu *et al*. (2011) conducted a cost analysis for different banana genotypes, and indicated that profitable markets existed in China for race 1- and TR4-resistant cultivars. Profitability of a given cultivar depended on whether plantations were infested with Foc, what race of Foc was found in infested fields, and market preferences. In infested soils, in which lower rents were charged but fewer cultivars could be grown, they recommended replacing susceptible cultivars with resistant cultivars or other crops.

The banana-breeding programs have faced enormous challenges (Ortiz & Swennen 2014). Primitive diploids that have been used as parents usually have poor agronomic and fruit traits, and introgression of disease resistances that they possess into advanced lines can take several generations. The polyploid nature of the crop; long generation times from planting to seed production; the large size of this plant and the corresponding need for large areas for hybrid evaluation; genetic abnormalities that exist in many parental lines; the need for final products to be parthenocarpic and sterile; and the low fertility of cultivars that need improvement are additional hurdles that impede progress (Ortiz & Swennen 2014).

Resistance to TR4 is found in several bred hybrids, especially those developed by the program at the Fundación Hondureña de Investigación Agrícola (FHIA) in Honduras. However, hybrids from FHIA and other programs possess only some of the agronomic, post-harvest and organoleptic standards that are found in the Cavendish clones. Somaclonal mutants, the so-called Giant Cavendish Tissue Culture Variants (GCTCV), have enhanced resistance to TR4, and several are now widely tested or used in Southeast Asia (Hwang & Ko 2004). Even though the somaclones are not completely resistant, can usually be grown for only one or two cycles in TR4-infested sites, have longer cycle times, and produce misshapen fingers and hands (Walduck & Daly 2007), they are currently the best alternatives for 'Grand Nain' and other Cavendish cultivars (Ploetz & Evans 2015).

Genetic transformation of banana has become relatively commonplace, and disease resistance is one of the most sought-after traits (Ortiz & Swennen 2014). There are convincing arguments for using genetic transformation to create resistant genotypes, especially when targets, such as Cavendish-like export bananas, are difficult to improve via conventional breeding (Aguilar Morán 2013). When and whether GMO (Genetically Modified Organism) bananas will be accepted in the marketplace is not clear. However, even if GMO bananas were accepted there are still significant technical challenges to creating cultivars that resist TR4 or other races of this pathogen in the field. In general, genes that provide durable resistance in this crop have neither been identified nor transferred to susceptible genotypes. Although a range of transgenes have been tested, only short-term results from greenhouse or incubator experiments are usually reported (Ploetz & Evans 2015). Long-term field results for GMO bananas are needed to demonstrate the promise they might offer when combatting this disease.

TR4 in the Jordan Valley

During September 2014, banana production areas in the Jordan Valley were surveyed for Panama disease; it was diagnosed in the following sequence (Stover 1962): 1) The oldest leaves of suspect plants were chlorotic, wilted and ultimately necrotic (Fig. 1A & B). 2) When the pseudostems of such plants were examined, they were noticeably harder and more difficult to cut than healthy plants. 3) The xylem of affected plants exhibited a characteristic reddish-brown discoloration, which ranged from discrete dots to large confluent areas (Fig. 1C & D). 4) In representative cases, samples were taken from symptomatic xylem for subsequent isolation and identification of the pathogen.

For step 4, dried vascular elements were surface disinfested for 15s in 70% ethanol, 2m in 10% household bleach, rinsed in sterile water, blotted dry on sterile paper towels and covered by molten (49 °C) half-strength potato dextrose agar amended with 100 mg L^{-1} streptomycin sulphate. Single-microconidium isolates were stored for future use on sterile filter paper. Vegetative compatibility tests for the isolates were conducted with nitrate-nonutilizing mutant testers for VCG 01213-01216, and the TR4 PCR diagnostic test of Dita *et al.* (2010) was used to confirm the identity of these isolates (Fig. 2).

West Bank of the Palestinian Authority. In 2011, 6,160 tons of bananas, valued at US$7 million, were produced on 200 ha in the Jericho region (FAOSTAT 2013). During the survey, virtually all of this production area was examined for Panama disease (see Fig. 3). Plantings of 'Dwarf Cavendish' and 'Grand Nain' were surveyed on the outskirts of Jericho, as well as in the Al-Dyouk Valley, the oldest and largest contiguous banana production area that remains in the West Bank and which is in close proximity to Jericho. No evidence for Panama disease was observed in any of the surveyed plantings. Notably, these production areas are in close proximity to the frontier with Jordan, and are connected via the Allenby Bridge (aka the King Hussein Bridge or Al-Karameh Bridge) to the TR4-infested Shooneh Janoobiyeh district in Jordan.

Israel About 1,000 growers and 2,000 casual workers are employed in the banana production sector in Israel. In 2011, 112,698 tons of fruit with a wholesale value of US$113 million were harvested from ca 2,500 ha (FAOSTAT 2013; Yair Israeli, personal communication). Production occurs in the Western Galilee (500 ha), Carmel Shore (1000 ha) and Jordan Valley (1,000 ha) from Kibbutz Sha'ar Hagolan in the north to Kibbutz Gesher in the south along the Jordan River and the Kineret. There is a small plantation south of Beit Shean in Moshav Revia and Mechola areas.

Banana production areas in the Jordan Valley, including those on the frontier with the Shooneh Shamaliyeh production area in Jordan, were surveyed. Production there was of 'Grand Nain'. No evidence for Panama disease was observed, nor was the disease recognized or reported by plantation managers. Based on interviews of producers, extension agents and researchers at the Jordan Valley Regional Agricultural Research Center and the Volcani Center in Bet Degan, the disease is also not known in other production areas in Israel. Importantly, those with considerable knowledge of banana production in Israel, as well as Panama disease and TR4 in other countries confirmed that the disease was absent in

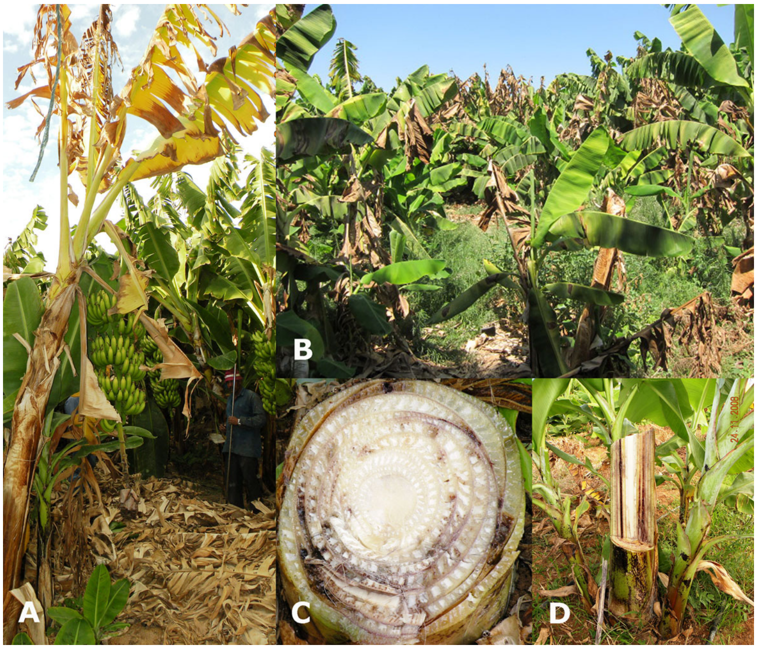

Fig. 1 Symptoms of Panama disease on Cavendish bananas in Jordan. External symptoms included: A) chlorosis and/or B) necrosis of leaves, progressing from the oldest to the youngest in a plant. Note in B) that buckling of leaves also occurred before they became chlorotic or necrotic. Internally, affected xylem and the associated parenchyma was reddish to dark brown which, when viewed in C) cross section was evident as discrete dots or contiguous sections of discoloration, and when viewed in D) longitudinal sections encompassed much of the pseudostem length

Israel (personal communications, Yair Israeli, Jordan Valley Banana Experiment Station, and Eli Khayat, RAHAN Meristem).

Jordan In 2011, 48,303 tons of banana valued at US$40 million were harvested in Jordan (FAOSTAT 2013). The area planted with banana increased from ca 1600 ha in 2006 to 2,090 ha in 2012, and production was of 'Grand Nain', 'Williams' and 'Baladi' ('Dwarf Cavendish'), all of which are Cavendish cultivars. Banana is cultivated in four areas: Shooneh Janoobiyeh (1500 ha), Dair Alla (2 ha), Shooneh Shamaliyeh (200 ha) and Ghor Safi (350 ha) (Fig. 3).

During the survey, Panama disease was not observed in the Shooneh Shamaliyeh production area immediately across the Jordan River and to the east of important banana production areas in Israel. Several cases of pseudostem heartrot, caused by *Fusarium* "moniliforme", were noted in the northern Shooneh Shamaliyeh, and producers were informed of the differences between this disease, which does not cause vascular streaking or systemic infection, and Panama disease, which does have these attributes (Stover 1972). However, symptoms of Panama disease were observed in two plantations in the mid- to southern portion of the Shooneh Shamaliyeh area (Fig. 3). A representative sample was confirmed as TR4

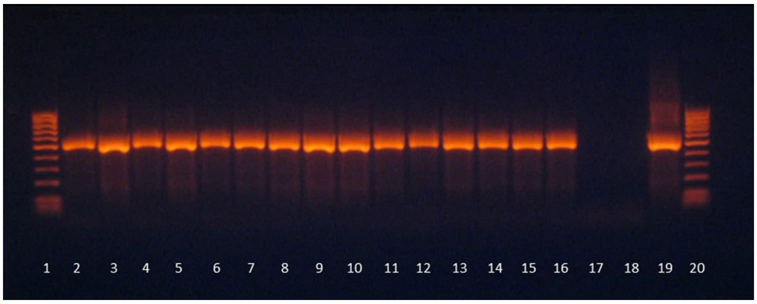

Fig. 2 PCR confirmation that isolates recovered from Cavendish plants affected by Panama disease during the September 2014 survey were of tropical race 4 (TR4), VCG 01213-01216. With PCR primers from Dita *et al*. (2010), diagnostic bands were generated for four isolates from sample #1 (lanes 2 – 5), six isolates from sample #2 (lanes 6 – 11), five isolates from sample #4 (lanes 12 – 16), and from a positive control isolate of TR4, CBS 102025 (lane 19). No DNA was amplified for an isolate from VCG 0125 (lane 17) or without template (lane 18). Fisher exACTGene 100 bp ladders are shown in lanes 1 and 20

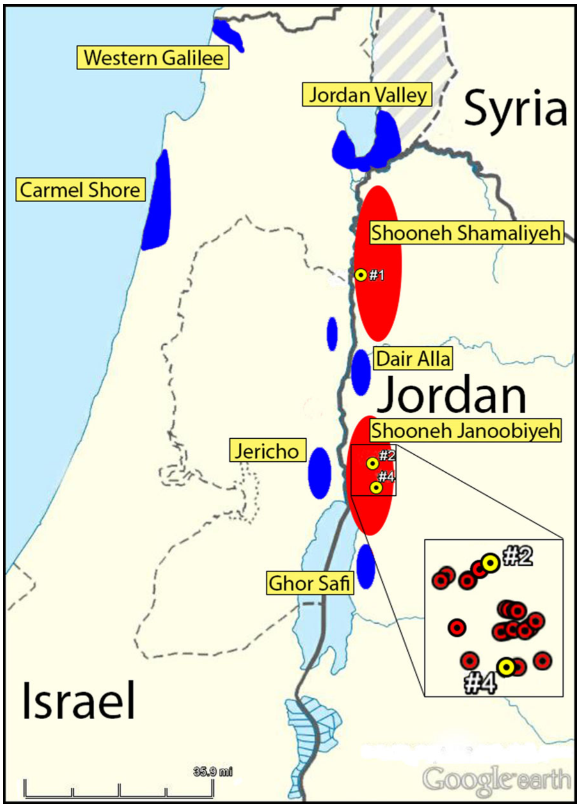

Fig. 3 Areas in the Jordan Valley that were surveyed for Panama disease and referred to in the text. Red production areas are those in which TR4 had been confirmed previously (Garcia *et al.* 2014) or in the present article, and blue areas are, as of this writing, free of the disease. #1, #2 and #4 refer to locations from which TR4 was diagnosed in the present study, and GPS locations for these samples and other locations where the disease was observed are indicated on the map and map inset

after VCG and DNA analyses (#1 in Fig. 3). This is an apparent, northward expansion of the distribution of TR4 in Jordan.

Symptoms were not observed in the Dair Alla area, but were prevalent in the Shooneh Janoobiyeh district. Producers in the district indicated that bananas had been lost to Panama disease since at least 2005, and that production was becoming exceedingly difficult. During the survey, many affected fields appeared to be abandoned and others that were in production had a large percentage of the plants/mats with symptoms (Fig. 1B). Representative samples were positive for TR4 (#2 and #4 in Fig. 3), corroborating a previous report of TR4 in the district (Garcia *et al*. 2014). Locations of symptomatic fields that were noted during and prior to the survey (August and September, 2014) are shown in Fig. 3, although they probably underrepresent how widespread the problem is in this area.

In general, it did not appear that fields in Shooneh Janoobiyeh, once affected, could be kept in production for more than 3 additional years. Abandoned, former banana fields were either left fallow or planted to other crops, and at least one producer had begun converting banana plantations to date palm production. There was an interest in how long the pathogen survived in affected fields, as some growers wanted to replant previously affected fields with banana. As noted above, this pathogen can survive for considerable periods in other production areas. Whether it could survive long under the extremely hot and arid conditions that are present in Jordan should be examined.

TR4 was assumed by many in Jordan to have arrived in their country from Israel. Tissue culture plants have been imported from Israel to Jordan, and since the initial outbreaks occurred in fields that were established with these materials Israel has been the presumed source of this outbreak. This is improbable, as only tissue culture plantlets have been imported from Israel into Jordan. Plantlets are easily infected when they are grown in infested soil, but they themselves are free of Foc and would, thus, not transmit the pathogen (Smith *et al*. 1998). Furthermore, there is no evidence for Panama disease in Israel. However, another species of *Fusarium* has been reported there as a fruit pathogen (Temkin-Gorodeiski & Chorin 1971). Unfortunately, Panama disease and *Fusarium* were confused by many during the survey ("If a *Fusarium* sp. has been reported on banana in Israel they must have Panama disease" was a typical assumption). In conclusion, TR4 was probably introduced to Jordan via other unknown sources.

The farms where Panama disease was first noted in the Shooneh Janoobiyeh district are no longer in production, and the original producer no longer grows banana. It was not possible to interview this producer regarding the original outbreak, nor was it possible to determine the origins of foreign workers in that or other plantings that were first affected by the disease. However, workers from Egypt were encountered during the survey, and movement of banana plants from Egypt to Jordan was indicated. Although TR4 has not been reported in Egypt, workers in that country have come from diverse locations in the Middle East. Clearly, discerning the avenues of TR4 dissemination in the Middle East would be an important first step in slowing its spread, as managing this problem is much more difficult once an area is infested.

A final, putative outbreak of Panama disease in Jordan was reported in Ghor Safi by Eli Khayat (RAHAN Meristem, personal communication). It could not be examined during the survey, but warrants investigation.

Summary and Recommendations

In summary, TR4 is apparently not present in Israel or the West Bank of the Palestinian Authority, but is established in two, or possibly three, banana-production areas in Jordan. Four situations were observed in the Shooneh Janoobiyeh district in Jordan:

i) In some new plantings there was no evidence of Panama disease and all plants appeared to be healthy. In these situations, harvest of at least one or more cycles of fruit would appear to be possible if the pathogen is not introduced during harvesting operations (see below).

ii) In a second situation, minor occurrences of Panama disease were observed with an inconsistent distribution in the plantation. Although spread from an outside source or development from a prior infestation was indicated in ii, the incidence of the disease was low enough to warrant containment to restrict the pathogen's movement (see Ploetz 2015). With rigorous care, it may be possible to continue production for two or more cycles in such situations.

iii) In a third situation, Panama disease was significant but restricted to the borders of plantings. In at least one of these situations it was determined that the affected border was adjacent to a former banana planting that was eliminated by Panama disease, suggesting that the pathogen had spread from the old to the newer planting. Ongoing production of fruit in these situations appeared doubtful, given the history and rapid development of the disease in this area.

In situations i, ii and iii, it is imperative that those who harvest fruit use clean, disinfested harvesting tools. Contaminated tools effectively disseminate the pathogen, and sanitation measures for tools that are similar to those instituted to manage the spread of Moko disease (see Stover 1972) should be considered here as they would assist efforts to slow the spread of TR4. Wholesalers who are contracted to harvest fruit should not be allowed to harvest without disinfesting their harvesting tools, and routine tool disinfestation should be conducted and monitored.

iv) In a fourth situation, plantings were severely affected and plants/mats exhibited symptoms in a more or less uniform distribution in the field (Fig. 1B). It was not clear whether such plantings were established in previously infested fields, or whether tissue culture plantlets that were infected in the nursery were used for establishment.

Under no circumstances should traditional seedpieces ("suckers") be used to propagate the crop; tissue-culture plantlets, which are widely available, should be used for new plantings. Until survival of Foc under Jordanian conditions is better understood (see below), it should be assumed that establishing new banana plantations in infested fields is risky. Regarding the possible infestation/infection of tissue-culture plantlets in nurseries, these facilities should use utmost care: soil from surrounding banana fields should never be used, irrigation water should be pathogen-free (either desalinized or from wells), and plants should never be in contact with nursery floors. Although one nursery with high production standards was observed in Shooneh Janoobiyeh, it was not clear whether others in the area observed similar standards. Work should be conducted on how long Foc could survive under the harsh local conditions in Jordan, as well as the weeds that might be reservoirs for the pathogen. It is conceivable that survival of the pathogen would be reduced in nonirrigated, fallow soil in which weeds were eliminated.

Management of Panama disease is largely restricted to excluding the pathogen from noninfested areas (Ploetz 2015). Where Foc is established, it is necessary to use clean production practices and resistant cultivars. As soon as possible, the GCTCV lines from Taiwan should be introduced from reputable, pathogen-free sources (i.e. the Bioversity International Transit Center in Leuven, Belgium) and tested in Jordan for performance against TR4 and market acceptance. Since fruit that are produced in Jordan are not exported, there could be a greater acceptance there of the slightly misshapen fruit that are produced by the GCTCV lines.

Unless strict quarantine measures are enforced, it is probable that TR4 will continue to spread in Jordan, thereby increasing the likelihood that it will eventually move to and become established in adjacent production areas in Israel, the West Bank and elsewhere in the Middle East. Managing established outbreaks of TR4 is virtually impossible. Even in the Northern Territory of

Australia, in which TR4 was initially isolated and well contained, TR4 has spread so widely that the Australian Banana Growers' Council (ABGC) recently announced the failure and withdrawal of quarantine restrictions on infested properties (Freshplaza 2014). Alarmingly, on 12 March 2015, TR4 was confirmed in northern Queensland in the center of Australia's primary banana-producing region (Freshplaza 2015).

A heightened awareness is needed throughout the Jordan Valley and elsewhere in the Middle East on how TR4 is disseminated and the damage it can cause. Spread of TR4 to new areas in the Middle East would have disastrous consequences for the production of this crop, especially in Egypt. Panama disease has been reported previously in Egypt on only 'Pisang Awak' (local synonym = 'Paradaica') (Ammar 2007), a cultivar that is highly susceptible to race 1 (Stover & Simmonds 1987). Other *Fusarium* spp. have been indicted as causes of pseudostem heartrot in Egypt on Cavendish cultivars such as 'Dwarf Cavendish' (= 'Hendi' and 'Basrii'), 'Williams' and 'Grand Nain', but Panama disease has not been reported on these cultivars (Ammar 2007; Ploetz 1994; Shalaby *et al*. 2007). Egyptian production, 95% of which comes from Cavendish cultivars, occurs mainly in the flood plain of the Nile River. Given the ability of Foc to disseminate in surface water and the use of water from the Nile to irrigate banana plantations, TR4 could spread rapidly and cause considerable damage if it were introduced to Egypt.

Better information is needed on how Foc would move the long distances that were implicated in the recent African and West Asian outbreaks of TR4. Although recent transcontinental jumps in its distribution may have resulted via avenues that are outlined above, other possibilities should be considered. In the early 2000s, the Australian government published a risk assessment for the importation of Cavendish fruit from the Philippines to Australia (Commonwealth of Australia 2004). It indicated that Foc could move as both symptomless infections of the vasculature of fruit crowns, and in pieces of infected leaf trash that would be associated with fruit shipments. Large quantities of Cavendish fruit are exported from Mindanao to the Middle East. For example, from 2008 to 2012, export to Jordan averaged 418,000 18 kg boxes year^{-1}, and substantial quantities of fruit were also exported to other banana-producing nations in the Middle East, including Egypt, Turkey, Pakistan, Oman, Bahrain, Syria and the United Arab Emirates (Republic of the Philippines 2012). Thus, there would be ample opportunities for the introduction of TR4 to the Middle East via this route. Better understandings of this potential means of spread are needed.

Conclusion

In conclusion, TR4 was reported recently in Jordan, Lebanon, Oman and Pakistan (Fusarium wilt of banana 2015; Garcia *et al*. 2014; Ordonez *et al*. 2015). Clearly, the pathogen has moved outside its former Southeast Asian confines and is established in the Middle East (Ploetz & Evans 2015). This signals a significant threat to Cavendish production in the region, which totals ca 3 million tons of fruit year^{-1} and is a major component of local agriculture economies. Without a heightened awareness of the threat that TR4 poses and the implementation of measures to impede its spread and impact, losses will increase, and Cavendish production will become increasingly difficult throughout the region.

Compliance with Ethical Standards The authors declare no conflicts of interest. The research did not involve Human Participants or Animals.

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