An analysis of the risk from *Xanthomonas campestris* pv. *musacearum* to banana cultivation in Eastern, Central and Southern Africa

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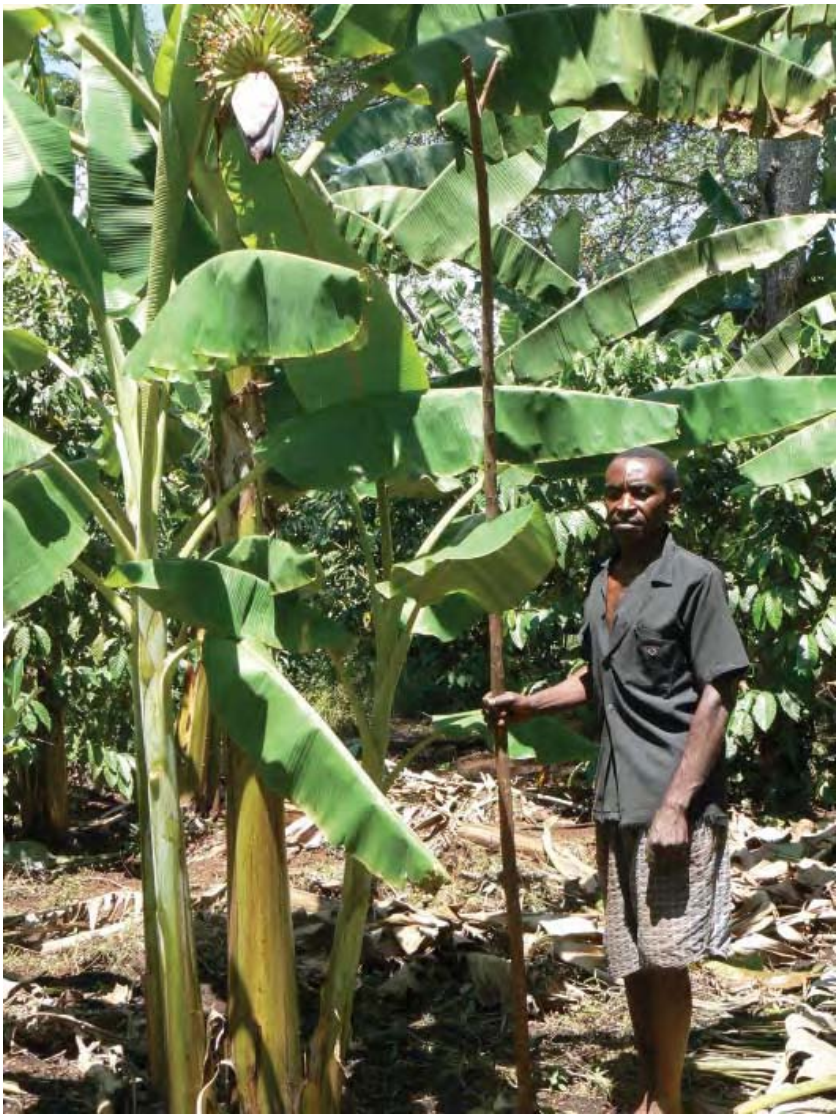

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Foreword

Xanthomonas wilt, which is commonly known in East Africa as banana Xanthomonas wilt (BXW), banana bacterial wilt or enset wilt, is a devastating disease caused by the bacterium *Xanthomonas campestris* pv. *musacearum*. It was first reported in 1968 in Ethiopia, where it remained confined until it was discovered simultaneously in 2001 in Central Uganda and the North Kivu province of the Democratic Republic of Congo (DRC). The subsequent spread of the bacterium throughout the Great Lakes region, where banana forms a large proportion of the diet for about 25 million people, is posing a serious threat to household food security and income.

In response to this situation, a number of national, regional and international organizations rated the disease a priority constraint and took steps to address the threat. The regional stakeholders, coordinated by Bioversity International and the FAO, developed a regional strategy based on multidisciplinary and multisectoral approaches.

The implementation of the regional strategy was facilitated when the Catholic Relief Services and the International Institute of Tropical Agriculture secured funds from USAID, as part of the Crop Crisis Control Project, to coordinate the fight against Xanthomonas wilt in Burundi, DRC, Kenya, Rwanda, Tanzania and Uganda. Bioversity International was sub-contracted to strengthen the capacity of key stakeholders in diagnosing and controling the disease on farm.

The need for a comprehensive pest risk analysis (PRA) to help policy makers in affected and at-risk countries was identified at a workshop entitled *Expert Consultation on Progressing the Road Map for the Control of Banana Xanthomonas Wilt in Uganda and across East Africa*, held in the UK in July 2006. The PRA will assist in the planning and execution of strategies for managing the disease, especially in threatened but disease-free areas.

The authors, scientists from the Central Science Laboratory in the UK and Bioversity International in Uganda, discuss the distribution and epidemiology of the causal agent and the measures farmers can use to protect their crops from contracting this deadly disease. They also analyse the impact of the measures deployed to date and make recommendations to reduce the risk of the disease spreading to other countries. It is hoped that the publication will help to raise awareness on the disease and stimulate investment for its control.

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1. Identity of pest

Scientific name
Xanthomonas campestris pv. *musacearum*

Synonyms
Xanthomonas musacearum

Taxonomic position
Bacteria; Proteobacteria; Gammaproteobacteria; Xanthomonadales; Xanthomona-daceae

Common name of the disease
Banana bacterial wilt, banana Xanthomonas wilt, enset wilt, Xanthomonas wilt of enset and banana. Local names used by farmers are also in existence e.g. Kiwotoka in Uganda.

Xanthomonas wilt is the common name recommended by the Committee of Common Names of Plant Diseases, International Society for Plant Pathology (Jones *et al.*, 2007).

Special note on taxonomy and nomenclature
Recent work based primarily on DNA sequence and fatty acid data has shown that strains of *X. campestris* pv. *musacearum* have very close homology to strains of *Xanthomonas vasicola* and most likely belong to this species. Accordingly, the name *X. vasicola* has been proposed for *X. campestris* pv. *musacearum* (Aritua *et al.*, 2006; 2007a), although this has not been formally approved as a new combination of names (Tindall *et al.*, 2006a; Young *et al.*, 2001).

The species *X. vasicola* is currently of single pathovar membership (pv. *holcicola*), comprising strains pathogenic to sorghum. However, recent studies by Vauterin *et al.* (1995; 2000) have reassigned to *X. vasicola*, at the species level, a number of *X. axonopodis* pv. *vasculorum* strains pathogenic to maize and sugarcane that were previously erroneously classified. Whereas the species name *X. vasicola* is accepted for the maize and sugarcane strains, the pathovar designation, pv. *vasculorum*, proposed by Vauterin *et al.* has not been approved (Young *et al.*, 2001). Thus, no formally accepted pathovar status is recognised for strains of *X. vasicola* from maize and sugarcane.

As a consequence of these suggested new memberships within *X. vasicola*, the pathovar status of strains therein requires revision. The limited pathogenicity studies undertaken to date have provided evidence for strains from enset and banana to be designated as *X. vasicola* pv. *musacearum*. In the study by Aritua *et al.* (2007a) a strain from banana was shown to elicit a pathogenic reaction on banana and maize (and, based on the taxonomy of the bacterium, possibly on sugarcane and sorghum also although this was not tested); whereas single strains of *X. vasicola* of sorghum and maize were only able to elicit disease on maize (Aritua *et al.*, 2007a). However, to confirm the pathovar status within *X. vasicola*, a more robust investigation encompassing a greater representation of strains of the putative pathovars on appropriate differential hosts needs to be undertaken.

2. Quarantine status

InterAfrican Phytosanitary Council (IAPSC)

X. campestris pv. *musacearum* is not included in the A1 or A2 lists of recommended quarantine pests of the IAPSC. Within the PRA area, Zanzibar is alone in placing restrictions on the import of banana to reduce the risk of Xanthomonas wilt gaining entry. *X. campestris* pv. *musacearum* has not been listed as a notifiable pathogen by any nation within the PRA area. No pest free areas are designated. The use of terminology such as endemic and outbreak is in common usage.

European and Mediterranean Plant Protection Organisation (EPPO)

X. campestris pv. *musacearum* has been proposed by France for inclusion in the European Union list of recommended quarantine pests, specifically with respect to France's overseas departments and regions of La Reunion, Guadeloupe, Martinique and French Guiana. This status is now under consideration by a working group of the European Food Safety Authority (EFSA).

3. PRA area

Xanthomonas wilt is currently restricted to Africa. It was first reported in Ethiopia by Yirgou and Bradbury (1968, 1974), although earlier records report a disease consistent with these symptoms as present in the 1930s (Castellani, 1939). Spread beyond Ethiopia was not reported until the disease was found in Uganda in 2001 (Tushemereirwe *et al.*, 2004). Subsequent spread to other countries of East Africa has proceeded relatively rapidly. In many instances, uncertainty exists as to the exact time of introduction, notably for those Central African countries affected by the disease.

The PRA area covers the following Eastern, Central and Southern African countries (the countries marked with an asterisk are members of the Banana Research Network for Eastern and Southern Africa): Angola, Botswana, Burundi*, Democratic Republic of Congo*, Eritrea*, Ethiopia*, Kenya*, Madagascar*, Malawi*, Mozambique*, Namibia, Rwanda*, Somalia, Sudan*, Swaziland, Republic of South Africa*, Tanzania*, Uganda*, Zambia and Zimbabwe.

Report of Xanthomonas wilt in the PRA area

Ethiopia

The first authenticated report of *X. campestris* pv. *musacearum* causing a wilt on Musaceae is that on enset in Ethiopia (Yirgou and Bradbury, 1968), with the disease reported as being present at many enset-growing localities in central and southern Ethiopia. However, a bacterial disease of enset with symptoms consistent with *X. campestris* pv. *musacearum*, was described previously in the late 1930s (Castellani, 1939). Subsequently, *X campestris* pv. *musacearum* as a pathogen of banana was first described in Kaffa province and later in warm, moist areas of other provinces (Yirgou and Bradbury, 1974). The occurrence of the disease on banana was observed to be more sporadic than on enset, but it was recognised that banana was a less common crop. It was recommended that 'great care should be taken to see that enset wilt does not escape and establish itself on banana in other parts of the world where it could pose a serious problem on this crop'. More recently, the disease has been reported as more common on banana than enset in western Ethiopia (Addis *et al.*, 2004).

Uganda

Xanthomonas wilt was first reported in Uganda in Mukono district of central Uganda in 2001 (Tushemereirwe *et al.*, 2004; Karamura *et al.*, 2006). By 2006, it was confirmed in 35 districts in all regions of the country. It has now been reported in 319 sub-counties out of a total of 986. It is said to be 'endemic' in 244 sub-counties with spread contained or controlled in 70 sub-counties (Tushemereirwe *et al.*, 2006; W. Tushemereirwe, KARI, Uganda, personal communication). The local name for the disease in Uganda is 'Kiwotoka' (Tushemereirwe *et al.*, 2006).

Democratic Republic of Congo

Xanthomonas wilt was said to have been first observed in 2001 on a few plants by farmers at Bwere Hill, Bashali Mokoto village in the Masisi district, which is 72 km northwest of Goma in North Kivu Province. However, it was not until January 2004 that agricultural officers first investigated reports of a serious new disease. Xanthomonas wilt of banana was confirmed in a 10 km radius around Bashali Mokoto village. The altitude in the affected area ranges between 1700 and 1740m, and cultivars grown include 'Kayinja' (90% of all banana plants), 'Sukari Ndizi', those in the East African Highland subgroup, the Cavendish subgroup and plantain subgroup. All were affected, but 'Kayinja' seemed the first to succumb and the Cavendish cultivars last. Later, a new disease outbreak was observed 20 km from the first, which lessened the chances of containment and eradication (Ndungo *et al.*, 2004; 2006).

Rwanda

Xanthomonas wilt was first officially observed in the northern part of the country in June 2005 and has been reported in the three districts of Cyanzarwe, Gisenyi and Kanama. Farmer reports suggest the disease may have been present as early as 2002 (Reeder *et al.*, 2007).

Tanzania

Xanthomonas wilt was first detected in September 2005 by farmers in Kabale village of Izigo Division within Muleba district, which is in the Kagera region bordering Lake Victoria, Uganda, Rwanda and Burundi. In January 2006, the outbreak was confirmed as Xanthomonas wilt and symptoms were also seen in nearby Kabale B sub-village. Plants in Izigo, Kikondo, Bumilo and Magata villages in Maruku district were also affected. The scattered outbreaks were separated from each other by tens of kilometres. It was estimated that banana plants belonging to about 100 households had the disease. Spread from eastern Rwanda was suspected (Mgenzi *et al.*, 2006a; 2006b), although there were no reported outbreaks in eastern Rwanda at the time.

Kenya

Xanthomonas wilt was reported in the Teso district of western Kenya, that borders Uganda, in September 2006 (Anon., 2006). Formal isolation and identification of the causal organism as *X. campestris* pv. *musacearum* has been reported (J. Smith, CSL, UK, personal communication).

Burundi

X. campestris pv. *musacearum* has been isolated from samples collected in Burundi (J. Smith, CSL, UK, personal communication). The samples had been collected during a survey undertaken on behalf of Catholic Relief Services (Anon, 2006).

4. Description of pathogen and disease

The full classical taxonomic description of the bacterium is provided by Bradbury (1986), with complementary fatty acid and molecular information available in Tushemereirwe *et al.* (2004) and Aritua *et al.* (2007a).

In summary: Gram negative, single, straight rod about 0.4-0.7 x 0.7-1.6µm, motile by a single polar flagellum. Chemo-organotrophic and obligately aerobic; without special structures or accumulations of poly-β-hydroxybutyrate or other storage products. Colonies on nutrient agar are yellow, smooth, circular and very mucoid. Growth is inhibited by 0.1% triphenyl tetrazolium chloride. Metabolic characteristics include: catalase positive; oxidase negative; do not denitrify or reduce nitrate; produce small amounts of acid from various carbohydrates and other carbon sources, but not from rhamnose, adonitol, sorbitol, dulcitol, meso-inositol, inulin or salicin. Gluconate is not immediately metabolised and asparagine is not used as a sole source of carbon and nitrogen simultaneously. DNA G + C content described at genus level ranges from 62.6 to 69.4%. Contains fatty acids 11:0 ISO, 11:0 ISO 3OH and 13:0 ISO 3OH that are unique to the *Xanthomonas* genus. Low levels of genetic variation have been observed between strains of *X. campestris* pv. *musacearum*, collected from the first outbreaks in Ethiopia and recent outbreaks in other countries (Aritua *et al.*, 2007). DNA sequencing studies have also shown that the species shares a very close relatedness to *X. vasicola* (Aritua *et al.*, 2007) (see Section 1).

Pathotype strain for *X. campestris* pv. *musacearum*: NCPPB 2005.
(Type strain for *X. vasicola* pv. *holcicola*: NCPPB 2417).

The disease causes loss both through death of the plant and rotting of edible/marketable fruit. External symptoms are characterised by a dull yellow wilt of the leaves, a drying rot and blackening of the male bud bracts and rachis, extending from the tip backwards along the rachis, and the premature and uneven ripening of fruit (Figure 1). Internal symptoms within the cut pseudostems are a yellow-orange discoloration of vascular tissue (vascular streaking) with some bacterial ooze and, within the fruit, a dark brown, soft rot (Figure 2). Symptom development is rapid under favourable conditions, and typically evident within -2-5 weeks under field conditions and 2-3 weeks under glasshouse conditions (Tripathi *et al.*, in press).

5. Isolation, identification and detection

Isolation of the bacterium from infected plant material is relatively straightforward provided the symptoms are not so advanced as to be dominated by secondary microbes (saprophytes). Isolation is recommended from the inflorescence stem. Excised plant material (2-5mm^3) should be removed aseptically from the internal region of an infected area (stem in cross-section) and placed in a small volume of sterile water (about 1ml) and left for 5 minutes for the bacterium to stream into the water. Isolation should be undertaken from the banana-water exudate by streaking onto a growth medium in a manner to obtain single bacterial colonies. A suitable medium for the primary isolation is NA medium, as this is a low-sugar-medium that will reduce excessive growth of competing, faster growing, saprophytes that might otherwise overgrow the *X. campestris* pv. *musacearum* colonies. Single colonies of *X. campestris* pv. *musacearum* will only be evident after 48 hours of incubation at 27-30°C and should be re-streaked to ensure purity on fresh media. A richer growth promoting media, such as YDCA that also buffers against acidification of the medium, is more suitable at this stage.

A semi-selective media for *X. campestris* pv. *musacearum* developed by Mwangi *et al.* (2007) is suitable for the isolation and quantification of the bacterium from non-sterile, non-plant backgrounds such as soil.

An array of diagnostic methods are available for the identification and detection of the causal organism, *X. campestris* pv. *musacearum*. Lelliott and Stead (1987) and Aritua, *et al.* (2007a) provide additional information on classical and modern approaches to identification. Recent first disease reports for Xanthomonas wilt have involved analysis of the bacterium fatty acid profile, DNA (*Gyrase*B gene sequence or rep-PCR profile) and pathogenicity on banana.

Diagnostic PCR (real-time and conventional) primers are also available (specific to *X. campestris* pv. *musacearum* and *X. vasicola* strains pathogenic to sorghum, maize and sugarcane) that have been based on sequencing of a rep-PCR fragment (Aritua *et al.*, 2007b). These primers are currently undergoing field validation in Uganda. It is anticipated that these primers will be useful in the identification, detection and monitoring of the pathogen in plant and non-plant environments.

6. Pest damage

The collation of robust yield loss data due to a disease of banana is inherently difficult within the farming systems of the Great Lakes region due to the informal and continuous cropping nature of production. The situation is further complicated by the mix of livelihood values associated with this crop that make any estimation of consequential impact hard to ascertain. Banana plants have market and subsistence values, both as a food crop and in the multiple non-food uses of the plant parts, notably the leaves and floral parts.

Limited work has been undertaken to quantify the economic losses due to bacterial infections of enset and banana in Ethiopia. In the report by Yirgou and Bradbury (1968), losses on enset were termed as heavy at many localities, and the subsequent report for banana observed 'Du Casse' as highly susceptible in Kaffa province (Yirgou and Bradbury, 1974) 'Du Casse' or 'Ducasse' is a synonym for 'Kayinja' (Jones, 2000).

In Uganda, the situation has been documented more thoroughly and the current distribution of the disease is well described (see Section 3). The most detailed survey on the impact of Xanthomonas wilt is reported in a Ugandan study led by Bioversity International (Karamura, 2006). This study estimated that between 2001 and 2004, the mean banana plot size decreased from 1.49 to 0.85 acres in areas where Xanthomonas wilt had been present for more than six months, while the mean farm size remained the same at 2.6 acres, with between 5 and 7% of banana farmers abandoning the crop. Yield levels were also reported as down by 65-80%, with associated income losses of 23-32% and reduced dietary intake from 290 to 205 kg/person/year. These findings have been used to estimate cumulative losses of up to US$4 billion if between 2001 and 2010 the disease incidence increased by 8% and 15% per year in East African Highland banana cultivars and 'Kayinja', respectively. The authors note that the actual rates of spread are much lower and that the calculated value does not take into account the coping strategies of farmers to mitigate the impact of Xanthomonas wilt.

7. Epidemiology of *X. campestris* pv. *musacearum*

Very little has been published in peer-reviewed literature on the epidemiology of *X. campestris* pv. *musacearum*, though the state of knowledge on this bacterium and the disease is rapidly progressing due to its importance. Numerous studies currently underway in Uganda and other countries will contribute to the knowledge base on the epidemiology of Xanthomonas wilt.

Host plants

Natural hosts of *X. campestris* pv. *musacearum* are cultivated enset (*Ensete ventricosum*) and banana (*Musa*) (Thwaites *et al*., 2000; Yirgou and Bradbury, 1968; 1974).

Among the *Musa* cultivars grown in the Great Lakes region of East Africa are those belonging to the East African Highland banana subgroup (AAA genome), which are used to make 'matooke' (steamed and mashed fruit) and beer. Also grown are the juice cultivar 'Kayinja' ('Pisang Awak' ABB genome), the dessert cultivars 'Sukari Ndizi' (AAB genome) and those of the Cavendish subgroup (AAA genome), and the cooking cultivars of the plantain subgroup (AAB genome).

Wild *Ensete ventricosum*, which is widely distributed in East and South Africa (Jones, 2000), is presumed to be susceptible, although no specific studies have been reported.

Both enset and banana belong to the Musaceae family, order, Zingiberales. Other family members of the order Zingiberales include: Cannaceae (Canna family), Costaceae (Costus family), Heliconiaceae (Heliconia family), Marantaceae (Prayer-plant family), Strelitziaceae (Birds of Paradise Flower family) and Zingiberaceae (Ginger family) (see http://plants.usda.gov).

Artificial inoculation studies have shown the ornamental *Canna orchoides* (Ashagari, 1985) and maize to be hosts (Aritua *et al*., 2007a); whereas a non-host status has been reported for peanut (*Arachis hypogaea*), bean (*Phaseolus vulgaris*), lucerne (*Medicago sativa*), broad bean (*Vicia faba*), wheat (*Triticum aestivum*), barley (*Hordeum vulgare*), sorghum (*Sorghum* sp.), maize (*Zea mays*), sweet pepper (*Capsicum annuum as C. frutescens*), tomato (*Lycopersicon esculentum*), tobacco (*Nicotiana tabacum*), aubergine (*Solanum melongena*), potato (*Solanum tuberosum*), sunflower (*Helianthus annuus*), lettuce (*Lactuca sativa*), castor oil (*Ricinis communis*), pelargonium (*Pelargonium* sp.), *Chenopodium album, Colocasia antiquorium, Commelina* sp., *Guizotia scabra, Kalanchoe quartinia, Snowdenia polystachya, Solanum nigrum* and *Tagetes minuta*. (Yirgou and Bradbury, 1974; Ashagari, 1985). Within these studies there is an inconsistency on the host status of maize.

The recently revealed taxonomic similarity of *X. campestris* pv. *musacearum* to *X. vasicola* presents a linkage to strains of known pathogenicity to sorghum, maize and sugarcane (Aritua *et al*., 2007a). As cited above, pathogenicity of *X. campestris* pv. *musacearum* on maize was shown (Aritua *et al*., 2007a). However, no specific tests for pathogenicity have been reported on sugarcane and sorghum and no studies have been undertake to investigate the presence of *X. campestris* pv. *musacearum* on maize, sugarcane and sorghum under field conditions.

The above pathogenicity assessments on *X. campestris* pv. *musacearum* have not provided a systematic analysis based on the taxonomy of known hosts or the bacterium. Critical gaps in knowledge exist with regards to pathogenicity on other mem-

bers of the Zingiberales family and the monocotyledons sugarcane and sorghum. It is noted that within the Zingiberales are a number of agricultural crops (e.g. ginger (*Zingiber officinale*)) and ornamental species, notably *Canna orchoides*, which has been reported as a host by artificial inoculation.

For the purpose of this PRA, the risk relating to host is limited to members of the Zingiberales and does not extend to maize, sugarcane and sorghum due to the preliminary nature of these data on pathogenicity. This area is identified as requiring further research in Section 14.

In planta translocation

X. campestris pv. *musacearum* is known to systemically invade all tissues of enset and banana after infection. This may involve the upward movement of bacteria through the vascular tissues if infection occurs in the lower parts of the plants (rhizome or pseudostem) or the downward movement of bacteria if infection occurs through the inflorescence (Ssekiwoko *et al.*, 2006; Blomme *et al.*, 2007).

The systematic nature of the bacterium is a highly significant factor in understanding the mechanisms of spread.

Host resistance/tolerance

Early field-based observations suggest all banana cultivars are susceptible to Xanthomonas wilt, but to varying extents. Initial research has now started to apportion, quantitatively in some studies, this variation to genetic, phenology and husbandry factors. Consideration on resistance (pathogen induced plant defence against the bacterium) versus tolerance (*in planta* basal level of pathogen supported without expression of disease symptoms) has not been investigated.

Genetic diversity amongst *Musa* species is well described and reasonably broad at the genus level (Ude *et al.*, 2002). However, amongst cultivated varieties, the gene pool is narrower, but still significant (Pillay *et al.*, 2006). Conversely, the genetic diversity amongst *X. campestris* pv. *musacearum* strains has been shown to be very narrow (Aritua *et al.*, 2007a). Thus, investigation for host resistance is somewhat simplified in that host/strain interactions are unlikely and any variation in disease expression recorded is most likely to be attributed to the host genotype when the method of inoculation circumvents husbandry and phenology factors (i.e. direct inoculation into the banana tissue).

Evidence in support of genetic resistance to *X. campestris* pv. *musacearum* is preliminary and inconclusive with respect to potential value for exploitation in breeding. Michael *et al.* (2006), in a field trial study on 40 local and exotic banana genotypes inoculated with a single *X. campestris* pv. *musacearum* strain, reported no genotypes as 'immune to infection'. All cultivars succumbed to Xanthomonas wilt. In this study, the different rates of disease progression, though not tested statistically, were assessed as not reflecting meaningful differences. However, in the study by Tripathi *et al.* (in press) statistically tested differences between cultivars in rates of disease progress and transitory symptoms are reported. The research by Tripathi *et al.* identified 'Pisang Awak' as the 'least resistant' and the species *Musa balbisiana* as the 'most resistant' to Xanthomonas wilt, with *M. balbisiana* exhibiting the capacity to recover. A con-

sistent reaction from each of 16 strains of *X. campestris* pv. *musacearum* tested was reported, supporting the contention that host/strain interactions are unlikely.

Variation due to resistance or tolerance factors has not been tested, especially in the context of transitory symptoms, but will be an important aspect in the epidemiology of the disease, especially where husbandry with knives is practiced. Potentially, tolerance may prove to be an exacerbating factor if it is shown that such cultivars are harbouring a bacterial load that, under favourable condition, manifests as disease or otherwise serves as a reservoir for wide-scale infection of other banana plants. Where plantations are mixed, with tolerant and less tolerant cultivars growing in close proximity, then it can be expected that Xanthomonas wilt will persist and cause yield losses. It is also evident from comparing these data that the authors are interpreting rates of symptom expression differently, as both studies show variation in rates for disease progress, but draw somewhat distinct conclusions. The need for a robust framework for evaluating host resistance was reported by Smith (2007).

In enset, transitory symptoms have been reported with cultivars 'Genticha' and 'Mazae' (Thwaites *et al.*, 2000), and cultivars 'Ado', 'Kembate', 'Hedesso', 'Soskila', 'Genticha' and 'Abate' have been reported to have 'relatively better tolerance than other cultivars' (Ashagari, 1985).

Saprophytic survival within the environment

Preliminary data on the longevity of inoculum in soil and plant debris is now becoming available, although robust conclusions cannot be drawn.

Experiments on survival of the bacterium in artificially inoculated soil and plant debris have been undertaken in Uganda. These studies demonstrated that *X. campestris* pv. *musacearum* populations declined rapidly in non-sterile soil, persisting for only 35 days, whereas in sterile soil, the population remained viable and detectable for 90 days of sampling. Survival in non-sterile soil was influenced by moisture content, with survival declining as moisture levels fell. Associated with plant debris, the bacterium survived for only 21 days when buried or on the soil surface, but for over 90 days when stored in the laboratory (Mwebaze *et al.*, 2006).

No research has been conducted on the causal factors for the reported decline in *X. campestris* pv. *musacearum* populations within non-sterile environments. Notably, the action of bacteria antagonistic to *X. campestris* pv. *musacearum* has not been explored and thus the potential to develop a biocontrol agent against Xanthomonas wilt is not known .

No research has been conducted on the survival of *X. campestris* pv. *musacearum* bacterium in water. The role of water in survival and spread of Xanthomonas wilt is not known.

On a cautionary note, these data on survival have been achieved through the use of semi-selective media and/or subsequent observed plant infection. Such methods have recognised limitations and currently no more sophisticated corroborative system for the detection (molecular or antibody based) of *X. campestris* pv. *musacearum* has been employed for monitoring populations within non-sterile environments. Molecular diagnostic tools have been recently developed (Aritua *et al.*, 2007b) and it is anticipated that supporting data will be available soon.

Modes of transmission

Airborne vectors

X. campestris pv. *musacearum* has been isolated from the nectar and from the ooze exuding through the cushions revealed by the fallen male flowers and the fresh openings made by the fallen bracts of both the male and female flowers (Tinzaara *et al.*, 2006). Airborne visitors pick up the bacteria when they visit the banana inflorescence. Twice as many insects visit the female flowers as compared to the male flowers. However, experiments suggest that infection occurs through the male flower cushions only, since no flower infection occurs when the male bud is removed, despite the presence of openings made by the fallen female bracts (Figure 3).

An inventory of insects visiting banana flowers in Uganda identified potential vectors. The most common insects were stingless bees (Family *Apidae*), fruit flies (Family *Drosophilidae*) and grass flies (Family *Chloropidae*). The bacterium was isolated from stingless bees, honeybees (*Apis mellifera*), fruit flies and grass flies that had been collected from male flowers of symptomatic and asymptomatic plants (Tinzaara *et al.*, 2006; Gold and Bandyopadhyay, 2005).

Nectar-sucking birds and bats are also frequent visitors to banana flowers and are suspected of transmitting the bacterium, but this has not been researched.

Use of contaminated husbandry tools

Many aspects of crop husbandry involve the use of a knife (panga). Recommended practices for the control of banana pests and diseases other than Xanthomonas wilt advocate the removal of senescing leaves and the removal of suckers to retain 3-4 pseudostems per mat. Similarly, harvesting of banana fruits and leaves is also undertaken by use of a knife. Spread of disease by contaminated knives is, therefore, a key mechanism by which the disease is spread locally (Eden-Green, 2006).

Transmission through movement of infected plant material

The systemic nature of the pathogen identifies the movement of plant material as a major pathway for dispersal. Accordingly, local and distant disease spread has been associated with the movement of plant suckers for replanting and also banana fruit, leaves (fresh and dried) and male buds (used as bungs for containers of banana beer) (Mwebaze *et al.*, 2006). The use of banana waste materials as mulch may also introduce the bacterium to new areas. However, no quantitative data have been obtained as to the frequency of infection within plant material (suckers and fruit) and evidence for dissemination in mulch, fresh and dried leaves and flower buds remains anecdotal. The recent availability of molecular diagnostic tools for the detection of the pathogen provides greater scope for this knowledge to be acquired.

Soil transmission

The role of soil transmission has not been studied, but based on the results of infection occurring through soil-borne inoculum (Mwangi *et al.*, 2006), bacterium longevity studies (Mwebaze *et al.*, 2006) and extent to which soil is carried within and between farms on lorries and machinery, animals and human feet, etc. inferences may be made that this method of dispersal is possible, but probably of a low significance for disease spread.

Animal activity and grazing

This has again not been studied, but theoretical pathways for spread, especially local spread, may be speculated. Animals that feed on the rhizome, such as the aardvark and porcupine, have been implicated in local spread in enset gardens of Ethiopia (Thwaites *et al*., 2000).

Factors affecting transmission

Host phenotype factors

Bracts: Banana cultivars with persistent bracts, have been observed to be less affected by Xanthomonas wilt in the field than those cultivars with dehiscent bracts (Temesgen and Handoro, 2004). This has been attributed to those cultivars with dehiscent bracts having exposed bract scars that, when fresh, exude sap that is attractive to insects and acts as an infection court for airborne vectors of the bacterium (see page 9). The persistent bract phenotype is seen as presenting a physical barrier to infection. However, this characteristic is not common amongst those cultivars preferred by farmers (Mwangi and Nakato, 2007).

Roots: Field trials have been undertaken in Uganda to estimate infection through the roots. Different infection rates were observed when infested soil was placed on the rhizomes of various cultivars. 'Enjagata' (AAA genome, East African Highland banana subgroup,) and 'Yangambi Km 5' (AAA genome) had less incidence of wilt than 'Gonja', 'Kibuzi' and 'Kayinja'. Generally, the more tolerant cultivars had fewer and shorter primary roots, and took an average of 69-77 days to wilt, while the more susceptible cultivars with a greater number of primary roots wilted within 40-50 days (Mwangi *et al*., 2006).

Climatic factors

Banana is a perennial crop that, whilst having seasonal peaks in productivity and husbandry requirements, is continuously cropped and thus under continuous husbandry. Therefore, it is probable that opportunity for infection and spread occur throughout the year, but vary according to the season. The seasonality of airborne vectors is not known, but is likely, especially for insects, to parallel patterns of rainfall that coincide with peak periods of banana growth and flowering. The risk of inflorescence infection will logically increase at these times.

It is known that altitude and land topography affect local environmental conditions, notably temperature and rainfall, that in turn influences suitability for growth of banana (with cultivar variation) and airborne vector populations, and this would be expected to influence disease spread. For example, male bud infection, which are mediated by airborne vectors, has not been observed in Ethiopia at altitudes over 1700 m (Addis *et al*., 2004) and reports from the Democratic Republic of Congo suggest that infection through the banana inflorescence is also not common above this altitude (Ndungo *et al*., 2006). Conversely, inflorescence infections are common around the lower elevations of Lake Victoria (altitude 1135m).

8. Introduction and spread

The mechanism for the entry of the pathogen from Ethiopia to Uganda is not known. However, noting the spatial separation of the banana cultivations of these countries and the probable absence of wild enset host within the intervening territory, it is most likely that this dispersal represents a long-distance outbreak as opposed to a progressive spread. Therefore, it is most probable that the outbreak in Uganda occurred due to the introduction of infected banana

material for agricultural purposes, although the movement of infected enset or other plant species of the Zingiberales, notably ornamentals or ginger, and even infected maize cannot be ruled out.

A different scenario exists for neighbouring countries with contiguous, or not remote, banana cultivation, as is typical amongst countries of the Great Lakes region. For these countries, the potential of introduction of Xanthomonas wilt is mainly through the local movement of infected banana material and also through airborne vectors.

Potential for entry

Data on production, import and export of bananas and plantains and the area under cultivation are available at the FAO Statistics Division (http://faostat.fao.org). The data show that most countries within the PRA area have land under banana cultivation and export and import banana fruit. However, these data should be interpreted with some caution as the informal and smallholder nature of the farming systems typical of these countries (with the exception of South Africa) makes accurate estimations difficult. Information on the movement of planting material, especially suckers produced by farmers and traded informally, is not captured within these data, although it is known that this represents a significant mechanism for disease spread. Similarly, the risk related to the movement of fresh banana leaves, as may be associated with packaging another traded commodity, is not known.

Information on cultivation and trade of enset and other members of the Zingiberales is not readily available. Information is available on the production and import and export of ginger for many of the PRA countries and this is significant for some countries. These data can be sourced at http://faostat.fao.org. However, the risk presented by the trade in ginger is undetermined as its host status with respect to Xanthomonas wilt is unknown.

Isolated outbreaks of the disease at a distance from known affected areas support the opinion that the pathogen is gaining entry over long distances and across international boundaries. The borders of many of the countries within the PRA area are known to be 'porous', with exchange of material through local trade being commonplace and largely unmonitored. The trade in banana fruit, and the use of suckers for planting and banana leaves for packaging are recognised as presenting a high risk of entry for this disease.

Potential for establishment and spread

Once it has entered an area, the pathogen is able to establish and cause disease rapidly provided the bacterium is received on a receptive infection court (wound, recently dehisced bract, etc.) in a viable state and the environment is conducive to infection and tissue colonisation. In regions suitable for banana cultivation, environmental conditions are likely to be conducive to disease development.

As described previously, mechanisms of spread include the husbandry practices of farmers, airborne vectors visiting the inflorescence, and the role of traders in the harvesting of banana fruit and transportation of banana products. The role of water, if any, is not known, although it has long been suspected that water may carry infection from upper to lower contour plantations.

In all countries recently affected by Xanthomonas wilt, rapid local spread has been evident. The appearance of the disease in countries bordering Uganda demonstrates how quickly the

pathogen can move through the intensive banana-growing areas around the Great Lakes region of East Africa. Knowledge of the susceptibility of banana cultivars suggests that the prevalence of highly susceptible cultivars within a region will impact on establishment and spread potential. It has been suggested that a high prevalence of susceptible cultivars, notably 'Kayinja' which is more prone to floral infection, will drive the spread of the disease (Smith, 2006; Mwangi and Nakato, 2007). A farmer's choice in the cultivars grown is known to be influenced by the local environment, notably the growing conditions and the marketing opportunities (Gaidashova *et al*., 2005). Accordingly, these factors will in turn influence the potential for disease spread.

It is beyond the scope of this analysis to determine to what extent the banana cultivation within and between the PRA countries forms contiguous cropping systems and whether high altitudinal factors, as appear to suppress disease spread, are likely to be a factor in reducing the rate of disease spread.

9. Potential for control using current practices, areas of uncertainty and future technologies

There has been much valuable work undertaken in Uganda to develop methods that farmers can use to reduce the risk of infection and in progressing with farmers' coping strategies for 'living with' the disease. Recommended practices have had to be cognisant of those control practices for other significant pests of banana, such as weevil, nematode and black leaf streak. Accordingly, the recommendations and insights below are ostensibly taken from experience gained from efforts to control the disease in Uganda and from knowledge of control practices for Moko bacterial wilt and bugtok caused by *Ralstonia solanacearum* and blood bacterial wilt caused by *Ralstonia* spp. (Eden-Green, 2006; Jones, 2000).

Cultural control

There has been much valuable work undertaken in Uganda to develop methods that farmers can use to reduce the risk of infection and in progressing with farmers' coping strategies for 'living with' the disease. Recommended practices have had to be cognisant of those control practices for other significant pests of banana, such as weevil, nematode and black leaf streak. Accordingly, the recommendations and insights below are ostensibly taken from experience gained from efforts to control the disease in Uganda and from knowledge of control practices for Moko bacterial wilt and bugtok caused by *Ralstonia solanacearum* and blood bacterial wilt caused by *Ralstonia* spp. (Eden-Green, 2006; Jones, 2000).

De-budding

The risk of infection through the inflorescence has been shown to be markedly reduced by the removal of the male bud by means of a forked stick (not cutting by a knife) as soon as possible after the last hand has been produced (Blomme *et al*., 2005; Ssekiwoko *et al*., 2006; Turyagyenda *et al*., 2006). This is preferred to bagging of the bunch until harvest, which achieves the same phyto-protective effect but is strongly disliked by farmers (E. Karamura, Bioversity-Uganda, personal communication). However, adoption of de-budding has been inconsistent amongst farming communities (Kagezi *et al*., 2006, Mwangi and Nakato, 2007), where farmers feel that de-budding affects the quality of the banana, especially for the juice cultivars (Bagamba *et al*., 2006).

Crop husbandry and harvesting

The role played by knifes in the spread of Xanthomonas wilt has been considered previously (see Section 9). It is not contentious that the sterilization of a knife with a chemical disinfectant between plants before pruning or harvesting will reduce the spread of disease. However, the implementation of this practice by smallholders and traders undertaking harvesting directly from farmers' fields is difficult as it involves additional effort in carrying disinfectant and water. Avoidance of plants with symptoms during routine husbandry and harvesting, leaving the management of these plants as a specific task, may reduce the risk of spreading the disease by knives. However, such a separation of tasks may also be unpopular with farmers and overlooks the fact that early or latent infection, or tolerant cultivars, will not present external disease symptoms as allows then to be managed as diseased (Smith 2007). These aspects have not been researched in detail.

A further husbandry practice that may contribute to disease spread is the use of banana trash derived from the farmer's own plants, neighbour's plants or from markets, as mulch. The extent to which banana waste may harbour inoculum and cause Xanthomonas wilt is not known. Accordingly, in areas where Xanthomonas wilt is found, the practice of mulching with banana plant materials should be discouraged, unless the mulch is known to be from a disease-free source (e.g. pest-free area).

Inspection and removal of infected plants

The identification and early removal of infected plants is seen as a key part of the control regime. Thus, the regular weekly inspection of plants by farmers, which allows for the early detection of infection and the timely implementation of a control practice, is important. This is not contentious, however, the approach taken for plant removal is open to debate. Plausibly, a fast acting herbicide treatment could be applied (see Chemical control, next page). However, such 'removal' of the infected plant from the smallholding or plantation is impractical for the majority of farmers. As a consequence the most widely advocated approach has been for infected plants to be dug up and cut *in situ* into small fragments to accelerate desiccation and decomposition. Yet even with this more pragmatic recommendation it is recognised that field situations are not easily managed and farmers are not inclined to remove an entire banana mat when only one pseudostem may be showing disease. It has further been observed that the work required for mat removal is high. Consequently the practice of mat removal is unpopular with farmers (Mwangi and Nakato, 2007). To add complexity, it is also noted that the chopping up of the pseudostem will release copious sap with a high pathogen inoculum into the environment (soil) and onto the cutting tools. When the fate of the bacterium in soil is not fully known and the practice of knife disinfection is not reliably practiced by farmers, the merits of this recommendation can be questioned. An argument could be made for a 'minimal intervention strategy', where only the infected inflorescence (if present) is removed to prevent airborne spread, isolating the infected plant mat to die naturally from the disease, whilst ensuring de-budding is thoroughly carried out amongst surrounding healthy plants.

Ssekiwoko *et al.* (2006) proposed a further option that, as a measure to save the mat, any pseudostem showing early symptoms of Xanthomonas wilt should be immediately removed at the base to minimise the risk of spread of the pathogen into the mat. The logic to this practice is only sound for floral infections (see Replanting strategies, next page).

The dynamics of *X. campestris* pv. *musacearum* populations during infection, disease expression and plant senescence, and with respect to persistence in soil and plant debris, has not been studied sufficiently to recommend an evidence-based approach to the safe removal of infected plants.

Chemical control

No commercial bactericide is available to protect or control infection, however, a herbicide treatment can be applied to kill the plant and by default kill the bacteria.

Research has shown that herbicides that kill infected plants relatively quickly can be used to reduce field inoculum. Field assessments have revealed that 2,4-D is more effective than glyphosate (Roundup®): pseudostems of 2,4-D treated plants start toppling after two weeks. It has been recommended that 1.6 ml of undiluted 2,4-D be injected into the pseudostem of an individual plant about 100 cm above the soil surface. Herbicide injection teams should comprise of one person making holes in the pseudostem with a metal rod while another person applies the herbicide with a syringe (Blomme e*t al.*, 2006).

However, the use of an herbicide to control Xanthomonas wilt within the PRA area is likely to be difficult to implement on a large-scale due to the informal nature of the cropping systems and the cost of the chemicals that are prohibitive if borne by the smallholders, without external assistance. Thus the use of an herbicide can only realistically be considered in the context of a limited action in address of a specific need. An example might be as a government-led contingency response to a new disease outbreak. In all cases, but particularly when advocating the use of herbicides to farmers who may be unfamiliar with the safety measures to take, supporting information on correct chemical application and disposal techniques should be provided.

No work has yet been undertaken on the use of chemicals to control insects on inflorescences, but such an approach is considered to be impractical and cost prohibitive.

As mentioned under cultural practices, chemicals can play a role in disinfecting pruning knives in crop husbandry and harvesting practices, but their accepted use by farmers remains a major challenge.

Replanting strategies

The importance of banana to households of the Great Lakes region places a high likelihood on farmers undertaking a replanting of banana after a Xanthomonas wilt outbreak. Consequently the demand for planting material has risen as a result of Xanthomonas wilt (Mwangi and Nakato, 2007).

Time required before replanting

Research on the longevity of the bacterium in soil is at a preliminary stage, but results to date suggest that only a relatively short fallow time period (a few months) may be required before replanting can be undertaken with a low risk of root infection (see Saprophytic survival under Section 7).

Accessing planting material

No significant formal supply of banana planting material is available within the countries currently affected by Xanthomonas wilt and consequently most planting material is sourced lo-

cally by farmers from their own or neighbours' fields. Mwangi and Nakato (2007) report 90% of suckers are sourced in this way. Traditionally, young suckers are taken from a mature mat and pared, with farmers selecting material from visually healthy plants. Allowing rhizomes to heal after paring and before planting has been shown to reduce risks of infection by *X. campestris* pv. *musacearum* (Mwangi *et al.*, 2006).

Research in Uganda has investigated the spatial dynamics of *in plant* infection with a view to safe-sucker use in replanting. It has been shown that when the bacterium enters the banana plant through the inflorescence, the bacterium must pass into the rhizome before the suckers become infected. This means that when the only symptom is shrivelled bracts, the rhizome is unaffected. This work suggests that the early removal of a flower-infected pseudostem at the mat base at the time of first floral symptoms can prevent spread of the bacterium into the mat (rhizome) and then into suckers (Ssekiwoko *et al.*, 2006; Blomme *et al.*, 2007). However, the same logic will not apply with infection introduced through husbandry practices or soil infection where leaf symptoms will be the first sign of disease and at this stage infection of the mat may probably have occurred (Smith, 2007).

Consideration has been given to the opportunity for private sector involvement in banana plant production by tissue culture and examples exist in Kenya and Uganda (Qaim, 1999). However, these initiatives do not meet the potential needs of farmers and have not been developed with assurances of pest-free status where tissue culture material is hardened on field nurseries prior to distribution to farmers. For wide scale use of tissue culture-derived plants by smallholders, strong demand needs to be realised through development of cultivars possessing advantageous traits desired by farmers and consumers, such as fruiting quality or resilience to pests, notably Xanthomonas wilt. It is noteworthy that 'Kayinja', which is particularly vulnerable to infection, remains popular with smallholders for particular purposes.

Preferred cultivars and prospects for breeding

As discussed previously, cultivars differ with respect to their predisposition to Xanthomonas wilt, which is a combination of resistance/tolerance, plant phenology and husbandry attributes, and no robust true resistance is known. Currently, no single banana cultivar is seen as presenting an elite combination of characteristics that might make it a preferred cultivar. Should a trait for tolerance/resistance be identified, its incorporation into locally preferred banana cultivars will not be accomplished easily or within a short timeframe by conventional breeding.

Recognising that conventional breeding of banana is problematic, notably due to sterility factors, research is ongoing in Uganda to produce a transgenic banana resistant to the pathogen. Banana tissue culture plants have been transformed with plant ferroxidin-like protein (*pflp*) and hypersensitive response assisting protein (*hrap*) genes isolated from sweet pepper that have been shown to function as bacteriocides in monocotyledons, such as rice (Tripathi *et al.*, 2006). However, transformed banana plants have still to be shown to be resistant to Xanthomonas wilt in glasshouse and field trials. Nevertheless, even if successful, this technology has a significant pathway of field efficacy, biosafety assessment and consumer acceptance to progress before release as a commercial cultivar.

10. Analysis of control

The critical component of the control effort in affected countries within the Great Lakes region has been the raising of awareness of the disease amongst rural populations that cultivate banana. Extensive media campaigns, including posters, billboards, radio and 'Going Public' (extension) events, have been undertaken where the disease has been most evident, notably in Uganda (Nankinga *et al.*, 2006). These campaigns have focused on the recognition of disease symptoms and the recommended cultural control practices to be taken. The effectiveness of these campaigns has been helped by the importance the people of the Great Lakes region place on banana, especially cooking banana, and by the disease having very striking symptoms that affect the main marketed product, the fruit. However, achieving awareness and effecting behavioural change amongst farming communities are not necessarily correlated. The importance people place on banana has also detracted from implementation of the control recommendations when these are destructive. Recommendations of mat destruction are not popular with farmers. Moreover, effecting behavioural change is even harder amongst farmers that have not witnessed the disease first hand.

Evidence for the effectiveness of the recommended practices is inconsistent and seemingly dependent of the willingness and ability of farmers and farmer groups to collectively implement recommendations (Tushemereirwe *et al.*, 2006). In the study by Mwangi and Nakato (2007) it was reported that the percentage of farmers practicing debudding varied according to country and that amongst the farmers differing levels of frequency in debudding were also observed. This study also reported that comparatively few farmers (in the order of 0-26%) were debudding as part of their response to control Xanthomonas wilt. The impact of just a few farmers that are either unwilling, or absent and therefore unable, to implement the control recommendations is seen as having significant negative consequences on the control of Xanthomonas wilt.

Understanding the differences in the management of 'Kayinja' and cultivars in the East African Highland banana subgroup, and how this pre-disposes the banana to infection, has proven an important aspect in appreciating the uptake by farmers of the control recommendations for Xanthomonas wilt. In explanation, 'Kayinja' is typically grown on smallholder homesteads in a poorly managed system with limited husbandry practices and where de-budding practices are uncommon. The flowers of 'Kayinja' are also sweeter and contain more nectar than the East African Highland banana cultivars, thus making visitation by potential airborne vectors of disease more numerous (Kagezi *et al.*, 2006). By contrast, East African Highland banana cultivars are predominantly grown on a larger scale and in commercial plantations. In such plantations, husbandry practices are more intensive, which presents a greater risk of mechanical spread of disease. However, routine de-budding is also more common and this reduces the chances of spread via the inflorescence. In the Kamuli district of Uganda, it has been estimated that the ratio of infection between 'Kayinja' and East African Highland cultivars is roughly 2:1 (Tushemereirwe *et al.*, 2006). The predisposition of 'Kayinja' to infection of the inflorescence may be the key factor responsible for the rapid spread of the disease (Mwangi and Nakato, 2007).

Nevertheless, positive examples of rehabilitation have been reported for plots of banana 'Kayinja' affected by Xanthomonas wilt in the Kiboga, Luwero and Mukono districts of Uganda after intensive extension activity, where participating farmers' records have shown an overall

80% decrease in infected plants with eradication reported on 50% of the farms. After rehabilitation, harvest levels recovered up to 80% of the pre-outbreak levels (Nankinga *et al*., 2006). Similarly, Mwangi and Nakato (2007) reported for central and eastern Uganda that 10.6% of framers had successfully eradicated Xanthomonas wilt from their farms over the past 2 years, and a further 18% of farmers claimed to have avoided the disease by implementing the control recommendations. In overview, there is some consensus that the current recommendations and awareness campaign has met with success, yet farmers in endemic zones are still experiencing losses and for many 'learning to live with the disease' best describes the current position (Mwangi and Nakato, 2007).

11. Management of outbreaks

The continuing frequency of outbreaks within affected countries is seen as the main threat to nationwide control (Smith, 2007).

Outbreak management requires a contingency response to be initiated that aims at containment and eradication. Foremost, for an outbreak to be managed, it would have to be found at a very early stage, before much secondary spread could have taken place, so that actions taken for eradication were practicable. An outbreak response could differ from that of a 'recommended control strategy for endemic zones' in that more intensive actions could be merited. For example, herbicides could be used and farmers compensated for razed banana plants, a measure that was part of the early effort to combat the spread of the disease in Uganda and Tanzania. However, experience has shown that herbicides are not used in outbreak management and the issues surrounding government compensation are complex, difficult to navigate and prohibitively expensive for resource poor countries to implement. Consequently, the experience within the PRA countries affected by Xanthomonas wilt has been that the advocated outbreak management response has not been substantially different to that for control. Follow-up activities of surveillance would be necessary to ensure that any eradication measures were successful.

If eradication is not possible, then containment is the next best control outcome. However, given the nature of spread and the uncertainties as to the actual means of long-distant dissemination to new areas this may be very difficult. It would require constant vigilance and policing to work, which may be beyond the resources of the countries concerned.

12. Prospects for continued exclusion

Countries in the PRA area for which Xanthomonas wilt is not reported and are therefore assumed free of the disease are Angola, Botswana, Eritrea, Madagascar, Malawi, Mozambique, Namibia, South Africa, Sudan, Somalia, Swaziland, Zambia and Zimbabwe. For these countries, immediate steps could be taken to prevent entry of the disease by restricting movement of banana and enset, and potentially other plant material of the Zingiberales, across international borders from locations where *X. campestris* pv. *musacearum* is established. However, noting the potential for latent infection, only countries where Xanthomonas wilt has never been recorded should be regarded as holding *X. campestris* pv. *musacearum*-free banana materials, and even amongst these countries evidence of effective surveillance for the disease

should be requested. Within the PRA area, it is not known if any of the countries reported as free of Xanthomonas wilt have set up the monitoring regimes needed to support that status and ensure the early detection of the disease and its causal organism should an outbreak occur. Moreover, it is known that where banana-growing areas are contiguous on both sides of an international border, exclusion will be highly problematic if one nation has the disease and perhaps impossible due to informal trade and the action of airborne vectors. The distribution and prevalence of wild enset is also not well known and may present a further pathway for disease spread. The lack of information of the contiguous nature of the banana, and to a lesser extent enset, along with potential topography and altitude boundaries that may limit spread (see Section 7), represents a significant gap in knowledge when assessing risk of spread and prospects for exclusion.

The spatial isolation of banana cultivation areas in Zanzibar, Madagascar and the costal region of Kenya affords opportunity for exclusion measures to be implemented. Protected areas of cultivation have the potential to be declared as having pest-free status. Zanzibar, which has some autonomy from mainland Tanzania, has taken measures to prevent the introduction of Xanthomonas wilt, although the current status and implementation of these measures is not known.

13. Conclusions

X. campestris pv. *musacearum* is a pathogen of banana and enset with the potential to cause significant damage in Eastern, Central and Southern Africa, and beyond. Now that the pathogen is established in a wide area of an important banana production region, the chances of further spread are much greater than when it was occurring only in Ethiopia. A significant factor in the spread of the disease has been the intensive and informal nature of banana cultivation within the Great Lakes region that has greatly increased the movement of banana material (fruit, suckers for planting and leaves) without oversight of the plant health authorities. It seems probable that the disease will continue to spread throughout the PRA area if measures to counter the threat are not taken.

Eradication of the disease within outbreak sites before it can spread further will require planning. The formulation of outbreak contingency plans for existing affected countries and countries where the pathogen is not known to occur and banana cultivation is important is deemed essential for successful eradication of a first outbreak. The contingency response to an outbreak should be more intensive to that advocated for controlling the disease in endemic areas in an attempt to eradicate the disease e.g. the use of herbicides may be justified under a outbreak situation.

Once the disease is introduced and established, experience has shown that it is very difficult to prevent it from spreading to nearby areas where banana cultivation is informal and contiguous, as in the Great Lakes region of East Africa.

It may be possible to protect isolated areas of production, such as Zanzibar, Madagascar and the coast of Kenya, as pest-free areas. However, the practicalities and costs of policing the movement of banana material from affected areas into these special zones has not been considered in this PRA. It may be beyond available resources of many countries.

A validated tool for the detection of *X. campestris* pv. *musacearum* is required to support research and phytosanitary activities. A prototype detection tool is now available for validation (Aritua *et al.*, 2007b). Competent laboratories for the rapid identification of *X. campestris* pv. *musacearum* need to be established throughout the PRA area.

Topological and altitudinal factors seem to influence the rate of spread of Xanthomonas wilt, with high altitude areas showing reduced rates of spread. This may be attributed to differences in airborne vectors of the disease, but this has not been established. The cooler temperatures of the higher areas will also have an effect on disease expression.

There is uncertainty as to whether plants with close phylogenic affinities to banana and enset (order of Zingiberales; e.g. the ornamentals *Canna* spp., *Heliconia* spp., and *Strelitzia* spp. and the crop, ginger) may be hosts of the bacterium. Plants and plant parts of these species originating in areas where the disease occurs should be treated as potential hosts until more definitive knowledge is acquired on host range.

Based on new taxonomic studies on *X. campestris* pv. *musacearum*, there is also uncertainty as to the host status of maize, sugarcane, sorghum and graminaceous weeds. These species may play a role in the epidemiology of the pathogen. If they are important hosts, trade in these crops between countries in the PRA area may significantly lessen the chance of success of any phytosanitary measure taken to mitigate the risk of spread.

Airborne vectors, notably insects, transmit the pathogen and this seems to be a significant mode of primary infection in 'Kayinja'. Insects are more active during seasonal rains and there appears to be more infection at this time.

A barrier to infection ('disease escape') from airborne mediated infection through avoidance of inflorescence infection is evident in banana cultivars with a persistent bract phenology that consequently have greater field resistance compared to cultivars that shed their bracts (bract dehiscence) and expose entry points for airborne vectors of the pathogen.

In addition to bract dehiscence, root phenology and genetic (tolerance and resistance) factors have been shown to influence the incidence of Xanthomonas wilt in the field. Scope for recommending particular cultivars for use by farmers has been shown, however, these choices do not relate well to those cultivars preferred by farmers and consumers.

Survival of *X. campestris* pv. *musacearum* on plant debris and in soil may be of a relatively short duration, but more research is needed for confirmation. Persistence of the bacterium in water is not known. A robust knowledge of persistence of *X. campestris* pv. *musacearum* in the environment is essential to make recommendations on the removal of infected plants and the length of the fallow period before replanting. The identification microbial antagonists may also provide opportunity to manipulate environments against the disease and/or identify microbes that can be developed as biocontrol agents.

The distinct husbandry practices associated with the cultivation of 'Kayinja' type and East African Highland cultivars result in different primary pathways of disease spread. Infection via the inflorescence is more prevalent in 'Kayinja' type cultivars because farmers do not usually

remove the male bud and these cultivars seem particularly attractive to insects. In well-managed plots of East African Highland cultivars, male buds are usually removed, however, the more intense husbandry of these cultivars increases the risk of infection as a result of the use of contaminated pruning tools.

The role of harvesting by traders and the practice of returning banana waste from markets as mulch in the spread of Xanthomonas wilt are not established.

The movement of the Xanthomonas wilt pathogen through fruit trade has not been established. It is highly probable that fruit will harbour populations of the pathogen and that this infection may not be apparent by visual external or internal (finger test) inspection.

In bringing together all the known human and environmental factors that affect the incidence and spread of Xanthomonas wilt, it may be possible to map high and low risk areas. A risk-map would help determine the best locations for the placement of disease surveillance resources.

Prospects for breeding cultivars that are resistant to Xanthomonas wilt and acceptable to consumers by conventional means are poor due to sterility factors in cultivated banana.

The promising early research of a transgenic cultivar resistant to Xanthomonas wilt presents an expedient route for introducing resistance into those cultivars more popular with farmers and consumers. However, any GM approach has to be progressed with the fullest consideration of biosafety and public acceptance of the technology in mind.

The current guideline for control, with the strong emphasis on raising awareness on the symptoms of Xanthomonas wilt amongst rural population and advocacy of de-budding as the main line for control, is showing success, especially where farmers act collectively to address the disease.

The value of involving grass-root organisations, such as NGOs, in galvanising a government response towards raising awareness amongst rural communities to Xanthomonas wilt has been shown.

14. Research required to reduce uncertainties

The research areas outlined below are mainly based on the workshop report by Smith (2007). The order presented is not in order of priority.

More laboratory and field-based research needs to be conducted to confirm the pathogen's longevity in soil and decaying plant debris under field conditions. This knowledge is critical to fine-tune the recommendations on the best approach for infected plant removal and the minimum time that land should be fallow before replanting.

The importance of banana-based mulch as a source of inoculum needs to be investigated, especially when these materials are by-products of market operations and are composites of many farmers' waste.

The role of harvesting and trading banana fruit in disease spread needs to be investigated. Is educating traders' in responsible harvest and market practices a complementary approach to farmer focused disease control?

The cause of outbreaks of Xanthomonas wilt is still not known. Outbreaks need to be investigated to determine their origin, i.e. movement of suckers or other banana material, such as infected fruit. The role, if any, of insects and birds/bats in long-distance spread needs to be investigated.

The diagnostic tool developed by Aritua *et al.* (2007) for the detection of *X. campestris* pv. *musacearum* needs to be validated so that this technology can be used to support research and phytosanitary activities.

Research is needed to assess the extent to which differences in the susceptibility of cultivar to Xanthomonas wilt in the field are attributable to resistance/tolerance, phenology and husbandry factors.

The epidemiological consequences of varietal tolerance (versus resistance) to Xanthomonas wilt needs to be understood in the context of disease persistence in the field and spread.

The consequence of differences in cultivar susceptibility and their spatial density and distribution within cropping systems need to be investigated more fully as drivers of disease spread.

More detailed knowledge on the relationship between insects visiting banana flowers and the pathogen would identify those insects specifically implicated in transfer. With knowledge of the major vectors involved, it may be possible to develop a control strategy that complements de-budding.

The influence of altitude on disease incidence and spread needs to be investigated, looking at the role of airborne vectors, temperature and rain on disease systems.

The reaction of other Zingiberales members, such as ginger (*Zingiber officinale*), *Canna* spp., *Heliconia* spp. and *Strelitzia* spp., to the bacterium should be substantiated.

Risk models and spatial mapping tools should be developed that utilise available epidemiological knowledge on disease spread to identify areas of banana production that are at high risk of infection. This would aid the advance deployment of resources for raising awareness.

The proposed taxonomic alignment of *X. campestris* pv. *musacearum* with *X. vasicola* of sorghum, maize and sugarcane opens up a hitherto unappreciated epidemiological dimension to the pathogen. Research is required to substantiate the host status of maize, sugarcane and sorghum, along with other related monocotyledon species. Can these species under field conditions harbour inoculum of *X. campestris* pv. *musacearum*?

For areas where banana cultivation is in decline because of Xanthomonas wilt and farmers are diversifying into other crops, the economic and social consequences of these changed practices

need to be assessed. Coping strategies that mitigate the impact of Xanthomonas wilt and reduce dependence on banana, while taking account demands on ecological services, need to be developed in partnership with rural populations.

15. Recommendations pertaining to mitigating risk

Consistent and increased use of phytosanitary terminology in defining the risk of Xanthomonas wilt at the national policy level will assist in presenting a common platform for communication within and between countries.

The IAPSC should consider declaring *X. campestris* pv. *musacearum* an A2 quarantine pest for Africa.

African countries without the pathogen and with a banana industry should consider declaring the pathogen an A1 quarantine pest and develop a contingency response to its entry.

Countries affected with Xanthomonas wilt may look to establish and maintain 'pest-free areas'.

A region-wide government mandated surveillance and monitoring regime for Xanthomonas wilt and its causal agent *X. campestris* pv. *musacearum* should be implemented in the PRA area, supported by a rapid diagnosis capability by laboratories designated as competent for disease identification. This may extend to the targeted monitoring of the movement of banana material across international borders.

Continued emphasis should be placed on raising awareness of the disease and recommended control practices amongst the rural populations through working with extension and grass-root organisations to facilitate early disease reporting and the implementation of a rapid and effective control response.

A continued investment in research is needed on knowledge gaps on Xanthomonas wilt and in support of new cropping systems by farmers that result due to a decline in banana production.

Within affected countries, methods for the control of Xanthomonas wilt in banana plots should continue to be refined based on new knowledge and disseminated widely amongst the rural communities.

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17. Figures

Photo: E. Boa

Photo: J. Smith

Figure 1. Yellow wilt of leaf (left) and shrivelled male bud and uneven fruit ripening (right).

Photo: J. Smith

Photo: G. Blomme

Figure 2. Rotting of the flesh is characteristic of the disease (left). Vascular streaking and ooze within cross-section of pseudostem (right).

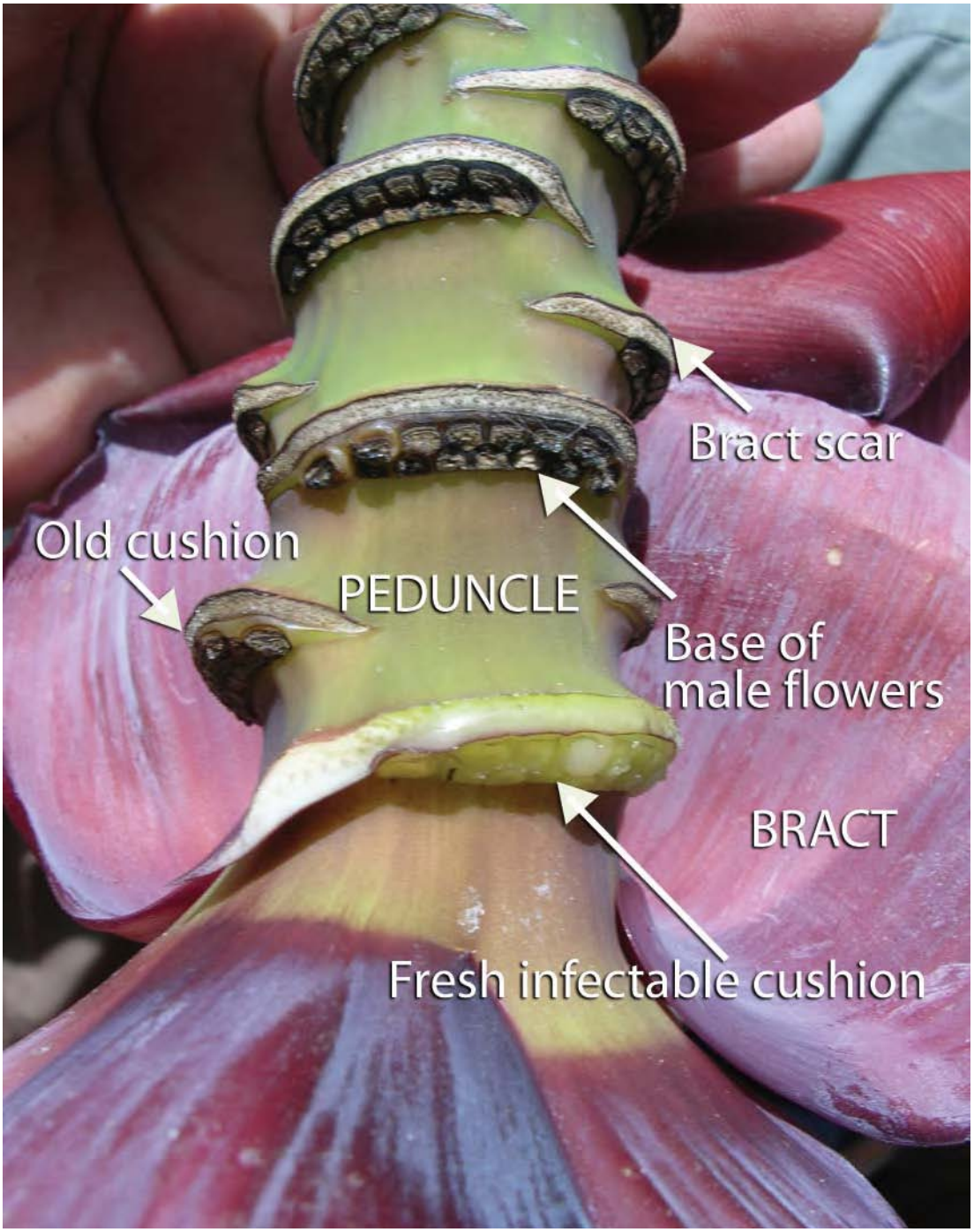

Photo: I. Buddenhagen

Figure 3. Parts of the inflorescence implicated in insect transmission.

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