Plant Pathology (2010) **59**, 403 Doi: 10.1111/j.1365-3059.2009.02124.x

Identification of *Xanthomonas vasicola* (formerly *X. campestris* pv. *musacearum*), causative organism of banana xanthomonas wilt, in Tanzania, Kenya and Burundi

B. A. Carter[a], R. Reeder[b], S. R. Mgenzi[c], Z. M. Kinyua[d], J. N. Mbaka[e], K. Doyle[f], V. Nakato[g], M. Mwangi[g], F. Beed[g], V. Aritua[ah], M. L. Lewis Ivey[i], S. A. Miller[i] and J. J. Smith[a]*

[a]*The Food and Environment Research Agency, Sand Hutton, York, YO41 1LZ;* [b]*Global Plant Clinic, CABI, Bakeham Lane, Egham, Surrey, TW20 9TY, UK;* [c]*Agricultural Research and Development Institute, P.O.Box 16, Bukoba, Tanzania;* [d]*Kenya Agricultural Research Institute, NARL Kabete, P.O. Box 14733-00800 Nairobi;* [e]*Kenya Agricultural Research Institute,Thika, P.O. Box 220, Thika, Kenya;* [f]*Catholic Relief Service, Avenue de Muyinga No. 5, Bujumbura, Burundi;* [g]*International Institute of Tropical Agriculture, P.O. Box 7878 Kampala;* [h]*National Agricultural Biotechnology Centre, National Agricultural Research Laboratories, P.O. Box 7065, Kampala, Uganda; and* [i]*The Ohio State University, Ohio Agricultural Research and Development Center, 1680 Madison Ave., Wooster, Ohio 44691, USA*

Xanthomonas campestris pv. *musacearum* (Xcm) causes a disease on banana (*Musa* spp.) and enset (*Ensete* spp.) known as banana xanthomonas wilt (BXW). Recent studies have shown that Xcm is a strain of *X. vasicola* (Aritua *et al*., 2008). However, the status of pathovars within the species remains unclear. Prior to its discovery in Uganda in October 2001, the disease had been limited to Ethiopia (first reported 1968). Since then the disease has spread to the Democratic Republic of Congo and Rwanda (observed in May 2004 and September 2005 respectively). BXW can cause high yield losses and is a high priority concern within the Great Lakes region. A comprehensive review of the pathogen and disease was recently published by Smith *et al*. (2008). Thus far, outbreaks in Tanzania, Kenya and Burundi have only been referred to in symposium proceedings and on various websites. These new records are thus officially reported here for the first time.

In Tanzania, the disease was first reported in the Kagera region of north west Tanzania, bordering Lake Victoria, Uganda, Rwanda and Burundi, in September 2005 (Mgenzi *et al*., 2006). Spread has continued, but not to other major banana growing areas. In Kenya, the disease was first reported in September 2006 in the Teso District, of western Kenya, bordering Uganda (Anon, 2006). Spread has since been reported as slow. In Burundi the disease was first observed during October 2006 (Anon., 2006). The current status of BXW in Burundi is unclear with no recent substantiated reports.

Bacterial cultures were isolated from diseased racemes from Tanzania and Burundi at CABI, UK and from Kenya at KARI (NARL). All cultures were identified to species level at FERA by fatty acid profiling (MIDI system) and DNA analysis using *X. vasicola* specific primers (Aritua *et al*., unpublished data) and partial sequencing of the *gyrase B* gene (Parkinson *et al*., 2007). Koch's postulates were fulfilled for all strains at FERA by stem inoculation of banana plants (height approximately 30 cm) with a bacterial suspension (200 µL with ~10^7 cfu/mL) under controlled environmental conditions (minimum temperature 27ºC). Identification of Xcm isolates from Burundi and Kenya was further supported by Ohio State University and KARI, respectively, using *X. vasicola* specific primers (Lewis-Levy Miller, unpublished data) that have a different target site to those of Aritua *et al*. (unpublished data).

Reference cultures are held by the UK National Collection of Plant Pathogenic Bacteria, Accession Nos. NCPPB 4392-5 (Tanzania), 4434 (Kenya) and 4433 (Burundi).

References

Anon. 2006. CRS/USAID/IITA Press Release. Bujumbura - October 31, 2006. Catholic Relief Services, Crop Crisis Control Project (C3P), http://www.reliefweb.int/library/documents/2006/crs-bdi-31oct.pdf

Aritua V, Parkinson N, Thwaites R *et al*., 2008. Characterization of the *Xanthomonas* sp. causing wilt of enset and banana and its proposed reclassification as a strain of *X. vasicola*. *Plant Pathology* 57, 170–7.

Mgenzi SRB, Muchunguzi D, Mutagwaba T, Mkondo F, Mohamed R, Aritua V, 2006. An out-break of banana bacterial wilt disease in Muleba district, Kagera region, Tanzania. AfricanCrops.Net [http://www.africancrops.net/News/april06/bananawilt.htm]

Parkinson N, Aritua V, Heeney J, Cowie C, Bew JD, Stead D, 2007. Phylogenetic analysis of *Xanthomonas* species by comparison of partial gyrase B gene sequences. *International Journal of Systematic and Evolutionary Microbiology* 57, 2881–7.

Smith JJ, Jones DR, Karamura E, Blomme G, Turyagyenda FL, 2008. An analysis of the risk from *Xanthomonas campestris* pv. *musacearum* to banana cultivation in Eastern, Central and Southern Africa. InfoMusa@ [http://www.promusa.org/index.php?option=com_content&task=view&id=66]

*E-mail: julian.j.smith@csl.gov.uk. *Accepted 6 April 2009 at http://www.bspp.org.uk/ndr where figures relating to this paper can be viewed.*

Plant Pathology (2010) **59**, 403–404 Doi: 10.1111/j.1365-3059.2009.02171.x

First report of *Xanthomonas hortorum* pv. *pelargonii* causing bacterial blight of geranium in Turkey

M. Mirik[a*], S. Unlu[b] and Y. Aysan[b]

[a]*Department of Plant Protection, Faculty of Agriculture, Namik Kemal University, TR-59030 Tekirdag; and* [b]*Department of Plant Protection, Faculty of Agriculture, Cukurova University, TR-01330 Adana, Turkey*

In the spring of 2006 and 2008 a serious outbreak of a suspected bacterial blight disease was observed on zonal geranium (*Pelargonium hortorum*) and ivy geranium (*P. peltatum*) plants grown for potted production in commercial greenhouses in the Turkish cities Adana, Mersin, Izmir and Istanbul. Disease symptoms on *P. hortorum* were V-shaped leaf yellowing, wilting, and stem necrosis but not leaf spots. The vascular system was discoloured, dark brown to black. However, the disease symptoms on *P. peltatum* were typically small round leaf spots or large, angular necrotic areas on leaf surfaces with stem necrosis but not wilting. Round leaf spots begin as small (2–5 mm), pale yel-

Plant Pathology

low, water-soaked areas on the underside of the leaf. Within two to three days, the spots become well defined, slightly sunken, and turn dark brown to black. All infected plant samples reacted positively with the ImmunoStrip test for *Xanthomonas hortorum* pv. *pelargonii* (Agdia). Isolations were made from leaf spots and discoloured vessels of the geranium plants on yeast dextrose calcium carbonate (YDC) agar. Twenty-one bacterial isolates from the diseased tissues formed yellow-coloured mucoid and convex colonies on King's medium B and YDC medium. All these isolates were characterized as non spore-forming, Gram negative, rod-shaped, motile, aerobic, oxidase-negative, catalase-positive and amylolytic-positive. Pathogenicity was confirmed by stab inoculation of healthy geranium cuttings with pure cultures of all the isolates (Nameth *et al.*, 1999). Reference strain GSPB 1955 and sterile distilled water were used as positive and negative controls respectively. Characteristic brown lesions at the point of inoculation were observed within five to seven days on geranium cuttings for all tested isolates. Identification of the isolates was confirmed by DAS-ELISA and by polymerase chain reaction amplification with pathovar-specific primers. A 1·2 kb fragment specific for *X. hortorum* pv. *pelargonii* (Manulis *et al.*, 1994) was obtained, as well as a 197 bp DNA product with the primer pair XcpM1/XcpM2 (Sulzinski *et al.*, 1996). All of the test results were similar to those of the reference strain GSPB 1955.

This is the first report of occurrence of bacterial blight disease caused by *X. hortorum* pv. *pelargonii* on geranium plants grown in commercial floriculture greenhouses in Turkey.

Acknowledgements

The study was supported by TUBITAK with the project number TOVAG 106O333. The reference strain GSPB 1955 was kindly provided by Dr. H. Batur-Michaelis, Georg-Agust University, Göttingen, Germany.

References

Manulis S, Valinsky L, Lichter A, Gabriel DW, 1994. Sensitive and specific detection of *Xanthomonas campestris* pv. *pelargonii* with DNA primers and probes identified by random amplified polymorphic DNA analysis. *Applied and Environmental Microbiology* 60, 4094–9.

Nameth ST, Daughtrey ML, Moorman GW, Sulzinski MA, 1999. Bacterial blight of geranium: a history of diagnostic challenges. *Plant Disease* 83, 204–12.

Sulzinski MA, Moorman GW, Schlagnhaufer B, Romaine CP, 1996. Characteristics of a PCR-based assay for in planta detection of *Xanthomonas campestris* pv. *pelargonii*. *Journal of Phytopathology* 144, 393–8.

*E-mail: mmirik@gmail.com.

Accepted 1 July 2009 at http://www.bspp.org.uk/ndr where figures relating to this paper can be viewed.

Plant Pathology (2010) **59**, 404 Doi: 10.1111/j.1365-3059.2009.02247.x

First report of the quarantine pathogen *Xanthomonas arboricola* pv. *pruni* on apricot and plum in Switzerland

J. F. Pothier[a], C. Pelludat[a], M. Bünter[a], M. Genini[b], J. Vogelsanger[a] and B. Duffy[a]*

[a]*Research Station Agroscope Changins-Wädenswil, Plant Protection Division, CH-8820 Wädenswil; and* [b]*Office Cantonal d'agro-écologie, CP 437, 1950 Châteauneuf-Sion, Switzerland*

Xanthomonas arboricola pv. *pruni* (Xap), cause of stone fruit bacterial spot on many *Prunus* species, is a European and Mediterranean Plant Protection Organisation (EPPO) A2-list quarantine pathogen (Anonymous, 2006). It can cause significant yield loss and tree death. In Europe, Xap occurs in France, Italy, Bulgaria, Romania and Ukraine, but appears to be spreading due to changes in trade patterns, climate, host range and/or pathogen genotype. This is the first report of the bacterial pathogen in Switzerland, occurring with very limited distribution.

Xap was first detected on apricot trees and fruit (*Prunus armeniaca*) showing symptoms in 2005 in four orchards located in canton Valais, the primary Swiss production region. It was detected in 2009 on Japanese apricot (*P. mume*) inter-cropped with apricot in two of these orchards. Isolation from symptom-bearing fruit, leaf and canker samples using YCDA medium confirmed recovered colonies to be Xap based on colony morphology, quinate metabolism, serology (Neogen Europe, EXPRESS™), *gyrB* sequencing (Parkinson *et al.*, 2009; GenBank Accession No. FJ719771 – FJ719773) and pathogenicity bioassays with detached peach leaves. Direct detection in symptom-bearing fruit, leaves and cankers was achieved in 100% of samples using Xap-specific PCR primers (Pagani, 2004) and in 40% using *X. fragariae*/*arboricola* primers (Weller *et al.*, 2007). These primer sets gave 100 and 93% agreement, respectively, with standard identification methods when applied to Xap isolates. All Swiss isolates were copper sensitive.

During intensive surveys between 2006 and 2009 throughout the region where the bacterium was first detected (208 ha), no incidence of disease has been observed outside the four original infected orchards. In three of these orchards Xap is considered eradicated, with all symptom-bearing trees removed in the fourth. This suggests probable introduction via planting material rather than fruit or aerosols. Xap is considered present with limited distribution but not eradicated.

Acknowledgement

This work was funded by the Swiss Secretariat for Education and Research within COST Action 873.

References

Anonymous, 2006. *Xanthomonas arboricola* pv. *pruni*. *EPPO Bulletin* 36, 129–33.

Pagani MC, 2004. *An ABC Transporter Protein and Molecular Diagnoses of* Xanthomonas arboricola *pv.* pruni *Causing Bacterial Spot of Stone Fruits*. Raleigh, NC, USA: North Carolina State University, PhD Thesis.

Parkinson N, Cowie C, Heeney J, Stead D, 2009. Phylogenetic structure of *Xanthomonas* determined by comparison of *gyrB* sequences. *International Journal of Systematic and Evolutionary Microbiology* 59, 264–74.

Weller SA, Beresford-Jones NJ, Hall J, Thwaites R, Parkinson N, Elphinstone JG, 2007. Detection of *Xanthomonas fragariae* and presumptive detection of *Xanthomonas arboricola* pv. *fragariae*, from strawberry leaves, by real-time PCR. *Journal of Microbiological Methods* 70, 379–83.

*E-mail: duffy@acw.admin.ch.

Accepted 28 October 2009 at http://www.bspp.org.uk/ndr where figures relating to this paper can be viewed.