DISEASE NOTES OR NEW RECORDS

First records of the papaya strain of *Papaya ringspot virus* (PRSV-P) in French Polynesia and the Cook Islands

R. I. Davis^{A,G}, L. Mu^B, N. Maireroa^C, W. J. Wigmore^C, M. Grisoni^D, M. F. Bateson^E and J. E. Thomas^F

APlant Protection Service, Secretariat of the Pacific Community (SPC), PMB, Suva, Fiji Islands.

BService du Développement Rural, Département de la Protection des Végétaux, BP 100, Papeete, French Polynesia.

CTotokoitu Research Station, Cook Islands Ministry of Agriculture (CIMoA), PO Box 96, Rarotonga, Cook Islands.

DCentre de Coopération Internationale en Recherche Agronomique pour le Développement, Pôle de Protection des Plantes,
Saint Pierre, 97410, La Réunion, France.

^ESchool of Life Sciences, Queensland University of Technology (QUT), GPO Box 2434, Brisbane, Qld 4000, Australia. ^FDepartment of Primary Industries and Fisheries (DPI&F), AFFS, Horticulture, 80 Meiers Road, Indooroopilly, Old 4068, Australia.

^GCorresponding author. Email: RichardD@spc.int

Abstract. The papaya strain of *Papaya ringspot virus* (PRSV-P), the cause of papaya ringspot disease, was confirmed in French Polynesia and the Cook Islands by double antibody sandwich enzyme linked immunosorbent assay (DAS-ELISA). In French Polynesia, the virus has probably been on the islands of Tahiti and Moorea for several years, but appears not to have spread to eight other islands. In contrast, PRSV-P has only recently appeared in the Cook Islands and is now the subject of an eradication campaign.

Papaya ringspot disease, caused by the papaya infecting strain of *Papaya ringspot virus* (PRSV-P; genus *Potyvirus*), is one of the most serious diseases of papaya in many countries of the world because of its effect on tree vigour and fruit set and quality (Gonsalves 1998). However, it is not widespread in the Australasia–Pacific region. PRSV-P was confirmed in Hawai'i in 1949 (Jensen 1949), in south-east Queensland in Australia in 1991 (Thomas and Dodman 1993), and in Saipan, Northern Mariana Islands and Guam in 1994 (Kiritani and Su 1999). In the Pacific Islands, papaya is an important fruit crop, of great value as an export commodity to some countries such as Fiji Islands and Cook Islands.

In French Polynesia, occasional observations of ringspot disease-like symptoms on papaya have been made since 1994 on the islands of Tahiti and Moorea. In late 2002, desiccated and surface-sterilised papaya leaf samples collected from Paea on Tahiti were sent to the Plant Virology laboratory of the Department of Primary Industries and Fisheries, Queensland, Australia under Australian Quarantine and Inspection Service (AQIS) permit No. 200205357. Here they were tested for PRSV by plate trapped antigen enzyme linked immunosorbent assay (PTA-ELISA) using antiserum supplied by S.-D. Yeh (University of Hawai'i) and gave positive results (absorbance readings greater than three times the mean of healthy control leaves). Following this,

fresh papaya leaf samples were collected from a number of locations on the islands of Tahiti, Moorea, Raiatea, Tubuai, Rurutu, Hiva Oa, Nuku Hiva, Tahuata, Ua Huka, and Ua Pou. They were tested in French Polynesia using a DAS-ELISA kit (Agdia Inc., Elkhart IN, USA). This survey returned nine more PRSV positive ELISA test results from five more locations on Tahiti and two on Moorea. No disease symptoms were observed on the other islands surveyed and 30 leaf samples collected from these islands tested negative for PRSV. The response in French Polynesia has been to employ quarantine measures to protect the outer islands and a strategy of destruction of infected plants followed by replanting with healthy seedlings on Tahiti.

On the island of Rarotonga in the Cook Islands, a single tree showing typical ringspot disease symptoms was reported to Cook Islands Ministry of Agriculture (CIMoA) in June 2004. As no other trees with symptoms were seen, this tree was immediately destroyed after leaf samples were collected and sent (surface sterilised and desiccated) to Fiji under Fiji Quarantine and Inspection Division permit No. 48681. Here, leaf tissue was triturated in 0.25 M phosphate buffer containing 0.1 M EDTA (pH 7.5) (PE buffer) as this is known to enhance ELISA test sensitivity for this virus (Gonsalves and Ishii 1980; Thomas and Dodman 1993). The leaf samples tested positive (absorbance readings greater than three times

Australasian Plant Pathology R. I. Davis et al.

the mean of four negative control leaves) by DAS-ELISA for PRSV using an Agdia antisera reagent set. A duplicate sample was sent to the Queensland University of Technology, Australia under AQIS permit no. 200319145. Here, they also tested positive for PRSV by reverse transcription polymerase chain reaction (RT-PCR). Sequence analysis of the PRSV coat protein-coding region indicated a close relationship (> 96.5% nucleotide sequence identity) with isolates of PRSV-P and W from Australia and Hawai'i (GenBank accession codes were PRSV-P Australia, U14736; PRSV-W Australia, U14739; PRSV-P, Hawai'i X67673).

126

A follow up survey of every commercial crop, plus many domestic properties on Rarotonga was conducted 5-6 weeks later to determine whether the virus was more widespread. The survey was timed to detect normal aphid-borne spread that may have occurred to nearby trees from the original tree prior to its destruction. Fresh leaf samples from 281 papaya trees were collected and tested for PRSV by ELISA on Rarotonga in the same way as described above for testing in Fiji. Seventy-one of these samples were from the affected plot (including the 55 nearest trees to the infected tree). A further 83 trees from within a 2 km radius of the disease outbreak were sampled and the remainder were taken from elsewhere on the island. No further positive results were obtained from this survey and CIMoA followed this by monitoring the area weekly. CIMoA is confident of achieving total eradication of this outbreak. Similar eradication success has been achieved before in Hawai'i in the 1960s (Gonsalves 1998).

The origin of both disease outbreaks is not known. There is a report from the Philippines that the virus is seed transmitted at the very low rate of 0.15% (Bayot et al. 1990) but these experimental results have not yet been repeated elsewhere. PRSV-P could move in infected papaya or cucurbit plants, though the latter is less likely as natural transmission from papaya to cucurbits is apparently rare (Bateson et al. 1994). However, legal entry of such plant material from countries where papaya ringspot occurs is not permitted in either French Polynesia or the Cook Islands. PRSV-P is very closely related to the watermelon strain of this virus (PRSV-W) which causes important disease in cucurbit crops through much of the world (Purcifull et al. 1984). PRSV coat protein gene sequence analyses suggest that elsewhere in the world PRSV-P has evolved by mutation from PRSV-W (Bateson et al. 1994, 2002; Tetsuo 2003). In south-east Queensland, Australia, it seems that it took over 20 years for PRSV-P to emerge after the first detection of PRSV-W (Thomas and Dodman 1993). In the Cook Islands, PRSV-W was known on cucurbit crops over 20 years before the outbreak reported here (Thomas 1980) and large-scale papaya production, often with cucurbit intercropping in recent years (N. Maireroa and W. Wigmore, unpublished data), has been a feature of horticultural production on Rarotonga throughout that time. At present, because of the close relationship between PRSV-P and PRSV-W isolates within the Australia-Pacific region, sequence analysis cannot confirm the origin of the outbreak in the Cook Islands. However, further analysis of PRSV-W isolates from the Cook Islands may help to determine if it arose by mutation from local PRSV-W isolates. With this in mind, a high level of vigilance for the appearance of any more diseased trees will be maintained in the Cook Islands.

Acknowledgements

This work was undertaken by the Secretariat of the Pacific Community's European Union funded Plant Protection in the Pacific Project. D. Persley, DPI&F, Queensland, Australia is gratefully thanked for helpful advice and guidance with the Cook Islands survey and response plan.

References

Bateson MF, Hendersen J, Chaleeprom W, Ha CV, Gibbs AJ, Dale JL (1994) Papaya ringspot potyvirus: isolate variability and the origin of PRSV type P (Australia). *Journal of General Virology* 75, 3547–3553.

Bateson MF, Lines RE, Revill P, Chaleeprom W, Ha CV, Gibbs AJ, Dale JL (2002) On the evolution and molecular epidemiology of the potyvirus *Papaya ringspot virus*. *Journal of General Virology* **83**, 2575–2585.

Bayot RG, Villegas VN, Magdalita PM, Jovellana MD, Espino TM, Exconde SB (1990) Seed transmissibility of Papaya ringspot virus. Philippines Journal of Crop Science 15, 107–111.

Gonsalves D (1998) Control of Papaya ringspot virus in papaya. Annual Review of Phytopathology **36**, 415–437. doi: 10.1146/ANNUREV.PHYTO.36.1.415

Gonsalves D, Ishii M (1980) Purification and serology of papaya ringspot virus. *Phytopathology* **70**, 1028–1032.

Jensen DD (1949) Papaya virus diseases with special reference to papaya ringspot. *Phytopathology* 39, 191–211.

Kiritani K, Su H-J (1999) Papaya ring spot, banana bunchy top, and citrus greening in the Asia and Pacific region: occurrence and control strategy. *Japan Agricultural Research Quarterly* 33, 22–30.

Purcifull DE, Edwardson JR, Hiebert E, Gonsalves D (1984) 'Papaya ringspot virus.' CMI/AAB Despriptions of Plant Viruses, No. 292. (CAB International, Wallingford, UK)

Tetsuo M (2003) Epidemiological and molecular analysis of papaya viruses in Asian countries. Food and Fertilizer Technology Centre Technical Bulletin 165.

Thomas JE, Dodman RL (1993) The first record of Papaya ringspot virus-type P from Australia. *Australasian Plant Pathology* 22, 2–7.

Thomas W (1980) Watermelon mosaic virus, the cause of a serious disease of cucurbits in the Cook Islands. *New Zealand Journal of Experimental Agriculture* **8**, 309–312.

Accepted 25 November 2004