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Quarantine in relation to Australian citrus imports and exports

Patricia Broadbent

Biological and Chemical Research Institute, NSW Agriculture, PMB 10, Rydalmere, New South Wales 2116

Abstract

The risks of introducing exotic citrus diseases through the importation of citrus fruit, seed and budwood are discussed. The experience with citrus bacterial spot in Florida has demonstrated that species identification and even pathogenicity testing are no longer adequate; serological and biochemical typing are essential before substantial eradication programs are commenced. A protocol for citrus budwood importations is discussed and an example given of the problems caused by accepting virus-free budwood from an 'approved' source. Allowing entry of viruses endemic to Australia could have disastrous consequences if more severe strains with a wider host range are introduced. The orange and mandarin stem pitting strains of citrus tristeza virus are examples given. Citrus relatives e.g. ornamentals such as *Murraya* can be hosts of canker, greening and citrus tristeza virus, and should undergo similar quarantine procedures to those for *Citrus* spp.

Introduction

Australian citrus industry Australia is the fourth largest citrus producing country in the Southern Hemisphere (after Brazil, Argentina and South Africa) and fifteenth in the world with 1.3% of total citrus production (1992 data from Table 157 of December 1994 Commodity Statistical Bulletin, Australian Bureau of Agricultural and Resource Economics). Plantings in Australia continue to expand slowly with 10.1 million trees in 1992–93 and 74% of these bearing (6 years and over) (from Table 151, Commodity Statistical Bulletin, 1994). There are approximately 2750 citrus growers (with a net annual return of over $20 000 per grower) in Australia. Total citrus production has averaged approximately 600 kt over the past five years. In 1992–93 it was 742 kt, of which 88 kt was exported (Tables 151 and 152, Commodity Statistical Bulletin, 1994). The gross value of citrus production in 1992–93 was approximately $290.5 million. This makes the citrus industry the second largest fruit industry in terms of gross value of production (1992 Agricultural Census of the Australian Bureau of Statistics). Valencia production is by far the largest citrus industry at 49% of total production, followed by navel production (32%). Mandarins, lemons and limes and grapefruit, although significant on a regional level, account for 12%, 5% and 3%, respectively, of total citrus production.

The volatility in the price of Brazilian frozen concentrated orange juice and the problems this has created for domestic producers of processing fruit, has resulted in a movement away from production of Valencia or processing oranges for concentrated orange juice toward navel oranges for fresh fruit, with increased interest in the fresh export market. Australian exports of fresh fruit to June 1994 totalled 100 897 tonnes. The main export markets have been South East Asia, Pacific countries, the Middle East and Europe. It has proved difficult to satisfy the requirements of the newly developed market in Japan for freedom from fruit fly and Fuller's rose weevil, while United States authorities had to be convinced that fruit from inland (Murrumbidgee Irrigation Areas, Sunraysia and Riverland) orchards posed no threat from Septoria spot and are free of fruit fly, black spot and lemon scab. To date only South Australia's Riverland region has exported fresh citrus to the United States of America.

Citrus imports into Australia comprise juice, fresh fruit and dried products. In 1993–94 juice

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imports totalled 8165 TSS (1 kg TSS = 10.3 L of single strength juice) valued at $16.1 million (preliminary data; Table 153, Commodity Statistical Bulletin, 1994). Fresh and dried fruit imports in 1993–94 are provisionally reported as 9396 t valued at $9.3 million. Orange, lemon and lime fruits are imported from the United States of America, grapefruit from the United States of America and Israel, and mandarins from New Zealand and the United States of America.

Trade and quarantine Quarantine is concerned with preventing the introduction and establishment of pests and diseases into new geographical areas (Hopper 1991; Navaratnam and Catley 1986). A pest is any form of plant or animal life or any pathogenic agent injurious or potentially injurious to plants or plant products (IPPC 1979). A quarantine pest is **a pest of potential economic importance to the area endangered thereby and not yet present there, or present but not widely distributed and being officially controlled** (Anon. 1993). The establishment of phytosanitary barriers to trade must now be justified under the Sanitary and Phytosanitary agreement of GATT. Assessment of the risks presented by trade in plant and plant products needs to be based on sound and structured biological principles of Pest Risk Analysis (Phillips *et al.* 1994).

This paper discusses those diseases exotic to Australia which would be of potential economic importance to the Australian citrus industry, if introduced.

Disease risks from fruit and seed imports

There are few citrus diseases which are exotic to Australia and which are carried on fruit and seed. The five of importance on fruit are the bacterial disease citrus canker (caused by *Xanthomonas campestris* pv. *citri* (Hasse, 1915) Dye, 1978), and the fungal diseases: mal secco (caused by *Deuterophoma tracheiphila* (Petri) Kanc. & Ghick), sour and sweet orange scabs (caused by *Elsinoe fawcettii* Bitancourt & Jenk. and *E. australis* Bitancourt and Jenkins) and Cercospora spot (caused by *Phaeoramularia angolensis* (Carvalho and Mendes) Kirk). It is not known if the slow growing strain of *Colletotrichum gloeosporioides* (Penz.) Sacc., which causes post-bloom fruit drop, is latent in citrus fruits.

Another fungal disease, black spot (caused by *Guignardia citricarpa* Kiely) causes serious losses in coastal orchards in New South Wales (Kiely 1949), but does not survive or cause symptoms in hot dry inland orchards (Barkley 1988). By contrast, black spot in South Africa was first reported in 1929 only in the cool misty areas of Natal, but in 1945 assumed more serious proportions when it spread to the hot dry subtropical East and North Transvaal (Wager 1952). Introduction to Australia of strains with a broader physiologic diversity could threaten export markets and reduce the viability of inland citrus. The International Plant Protection Convention (IPPC) definition of a quarantine pest does not take into account such physiologic specialisation or variation. Very well defined international differentials need to be known for a given pest and the pathogen variants need to be stable entities which the importing country can prove that it does not have (Phillips and Chandrashekar 1992).

Virus diseases pose almost no risk through fruit or seed imports because with the exception of psorosis virus in *Poncirus trifoliata* (L.) Raf. and citrange (*P. trifoliata x Citrus sinensis* (L.) Osb.) (Childs and Johnson 1966; Bridges *et al.* 1965), viruses are not transmitted through citrus seed.

All imported citrus seeds are treated with a bacteriocide (8-hydroxyquinoline sulphate).

Canker

What is citrus canker? Citrus canker is a generic term that includes all diseases of citrus caused by strains of *Xanthomonas* (Stall and Civerolo 1991). Citrus canker is generally divided into the following diseases: citrus canker A (Asiatic or true canker), widespread and serious in South East Asia and affecting a wide range of rutaceous hosts; cancrosis B (false canker) found principally in lemons in Argentina and Uruguay; canker C (Mexican lime cancrosis) in Brazil on *C. aurantifolia* (Christen) Swingle; citrus bacteriosis (D) in Mexico and citrus bacterial spot (E) in Florida, United States of America. The major differences in these diseases are the range of susceptible hosts among the

Rutaceae and the severity of the diseases (Stall and Civerolo 1991). Canker A, B, C and D are characterised by conspicuous erumpent lesions that develop on leaves, twigs and fruit. Severe infections result in defoliation, dieback, blemished fruit, and premature fruit drop. The causal bacteria were all designated as members of *X. campestris* based on physiological tests and they were classified as strains of *X. c.* pv. *citri* because of their pathogenicity to citrus. By contrast, citrus bacterial spot (CBS) is a nursery disease principally affecting the rootstock Swingle citrumelo (*P. trifoliata* x *C. paradisi* Macf.). Leaf lesions are flat with a necrotic centre, a water-soaked margin and sometimes a chlorotic halo. Fruit and orchard trees are rarely affected (Graham and Gottwald 1990).

The 1990 quarantine procedure for *X. campestris* pv. *citri* issued by the European and Mediterranean Plant Protection Organisation (EPPO) does not distinguish forms of citrus canker. EPPO recommends that 'citrus-growing countries prohibit the importation of planting material from countries where the disease occurs: they may also prohibit the importation of fruit from such countries' (Anon. 1990).

Arguments about the taxonomic identity and danger of the citrus bacterial spot or the 'nursery strains of citrus canker' have raged in the United States of America at every level from the Florida citrus groves to the Federal Courts.

The nursery strains are comprised of at least two and possibly three relatively distinct groups on the basis of pathogenicity (Graham and Gottwald 1990). In contrast to Asiatic citrus canker A strains that share a common epitope as revealed with a monoclonal antibody (mAb) A_1, xanthomonads associated with citrus bacterial spot were antigenically heterogeneous and shared epitopes with other pathovars of *X. campestris* (Alvarez *et al.* 1990).

Vauterin *et al.* (1991) compared 61 strains belonging to all five described pathogenicity groups of *X. campestris* pv. *citri* (i.e. A, B, C, D, and E) and representing a broad geographical diversity. All pathogenicity groups were related to each other by a DNA binding degree of over 60%, indicating that they all belong to one species. The results of Vauterin *et al.* (1991) do not confirm the reclassification of the pathogens in *X. campestris* from citrus into two separate species (Gabriel *et al.* 1989) but the pathogenicity groups A and E could be clearly delineated.

Florida eradicated more than 20 million trees in more than 100 nursery and orchard locations at a cost of $94 million (Graham and Gottwald 1991). When it was shown that the bacteria causing CBS and canker A were different, the State of Florida faced substantial lawsuits from as many as 490 nurserymen (Longman 1989).

From the foregoing it is obvious that traditional taxonomic studies, pathogenicity tests, serological typing and the newer generation of molecular biological tests should be used together in pest risk analysis (PRA), especially where fruit imports or substantial eradication programs are involved.

Canker outbreaks in Australia Australia has had five outbreaks of citrus canker: in the Northern Territory in 1912 (Hill 1918), on Home Island in the Cocos (Keeling) Islands in 1981 and Christmas Island (Shivas 1987), on Thursday Island in 1984 (Jones *et al.* 1989) and near Darwin in 1991 and again in 1993 (Broadbent *et al.* 1992, Broadbent *et al.* 1995). The early Northern Territory outbreak of canker probably originated from Japan or China, as citrus trees were imported into Darwin from these sources and nearly every ship trading between these countries and Australia landed small consignments of citrus fruits at Darwin. Three Commonwealth Proclamations were issued in 1915 and 1916 and prohibited (a) citrus tree and fruit imports into Australia (but exempted citrus trees from the States of Arizona and California) and (b) the removal of citrus trees and fruits from the Northern Territory to any other part of Australia.

Although canker was detected in the Northern Territory in 1912 (Hill 1918), it was not confirmed until 1916 by Mr C C Brittlebank of the Victorian Department of Agriculture and by Prof H E Stephens of Florida. As far as possible every citrus tree in the Territory was carefully examined for canker by Hill in 1918. All diseased trees and 'canker-free' trees in proximity to canker-infected trees were destroyed. Also, all lands in the vicinity of areas where canker had been detected were placed under quarantine for a period of 5 years. This eradication campaign was unsuccessful and the decision was taken in 1922 to destroy all citrus trees in the Northern Territory, but this was later modified to exclude all trees south of the nineteenth parallel. It was not until 1925 that permission was granted to grow citrus again in the Northern Territory. No symptoms of canker were found during a survey in 1950 in regions that were featured in the original surveys (Mertin 1952).

No further outbreaks of canker were detected in Australia until 1981 when the disease was found on West Indian limes on Home Island in the Cocos Islands (Shivas 1987). In 1984, citrus canker A was detected on three West Indian lime and two sweet orange trees in gardens on Thursday Island in the Torres Strait (Jones *et al.* 1989) and on Christmas Island (Shivas 1987). Four additional infected trees were found on Thursday Island in 1985. By September 1988, Department of Primary Industries and Energy considered the outbreak on Thursday Island to have been eradicated (Catley 1988; Jones 1991). No further evidence of canker was found in a survey of citrus in the Torres Strait Islands carried out by Broadbent in 1990, as part of the Northern Australia Quarantine Strategy (NAQS).

Symptoms of citrus canker were detected on leaves, twigs and fruits of pummelos (*C. grandis* (L.) Osb.) in a young orchard at Lambell's Lagoon near Darwin in April 1991, during a NAQS survey. The causal agent was identified as *X. c. citri* (Asiatic or A Group) by pathogenicity reactions in various citrus hosts, fatty acid profiles and DNA fingerprints (Broadbent *et al.* 1992). The affected block of citrus trees was eradicated. No disease symptoms were observed at Lambell's Lagoon in the wet season of 1991–92, but canker A was again diagnosed in May 1993 (Broadbent *et al.* 1995).

Canker A will continue to be a threat to Australian citriculture. Indeed the risks may be increasing as travel between Australia and canker-infested Asian and Pacific Island countries grows and migration increases. The canker A disease could be expected to develop and spread rapidly, once the bacterium is introduced into a suitable environment, provided by susceptible citrus cultivars growing predominantly in coastal orchards and nurseries in Northern and Eastern Australia (Broadbent 1992). Rapid detection and strain identification of the pathogen and containment of any outbreak by prompt eradication of canker-infected trees are essential.

Mal secco

Mal secco (caused by *Deuterophoma tracheiphila*) is a tracheomycotic disease causing wilt and dieback of citrus trees, especially lemon and citron, in Israel and Mediterranean and Black Sea countries. Fruit on diseased branches are readily invaded and may show some vascular discoloration without external lesions. Under favourable conditions, normally tolerant citrus cultivars (e.g. grapefruit) will succumb to mal secco (Solel and Oren 1975). Control measures to date are unsatisfactory. The disease is highly destructive and would probably cause severe tree losses in Australian citrus growing areas. Care should be taken in the importation of fruit and budwood (especially lemons and grapefruit) from affected regions. Illegal importations of budwood from Italy and other Mediterranean countries are of particular concern.

Scab

Citrus scab diseases occur in most humid areas of the world and produce serious fruit blemishes which reduce the external quality of fruit for the fresh market. Although these diseases do not generally affect yields of citrus trees, they cost millions of dollars each year in downgrading fruit.

Sweet orange scab (caused by *Elsinoe australis*) is limited to South America (and perhaps Sicily) and causes serious fruit blemishes on sweet oranges and mandarins. Sour orange scab (caused by *E. fawcettii*) is widely distributed in Asia, South Africa, the South Pacific, the Gulf States of the United States of America, the West Indies and South America. In Florida, scab is most serious in commercial groves of grapefruit, Murcott tangor, Temple orange, as well as tangelo and other mandarin hybrids. Two biotypes of *E. fawcettii* occur in Florida. One biotype is most severe on rough lemon, grapefruit, Murcott tangor, sour orange and will even attack sweet orange fruit. Another biotype also attacks all of the above except sour orange, Temple orange and sweet oranges (Whiteside 1978). In Australia, by contrast, lemon or Tryon's scab caused by *Sphaceloma fawcettii* var. *scabiosa* is a commercial problem only on lemon fruits and rough lemon stocks in coastal areas.

Recent research in Australia indicates that *E. australis*, *E. fawcettii* and *S. fawcettii* var. *scabiosa* cannot be differentiated on the basis of colony morphology, spore size or shape (M. Priest, personal communication). Detached leaf assays were used to compare Australian, Florida and Argentine scab isolates on rough lemon, sour orange and grapefruit leaves. Australian isolates produced strong reactions only on rough lemon. One biotype of *E. fawcettii* from Florida produced strong reactions on all three species, whereas the second biotype reacted strongly only on rough lemon and grapefruit. None of the isolates of *E. australis* produced symptoms on leaves of any species (Timmer and Broadbent 1995).

Nucleic acid analyses indicated that isolates of *E. australis* could be readily distinguished from all other scab isolates by restriction analysis of the amplified internal transcribed spacer (ITS). ITS restriction patterns analysed for the biotypes of *E. fawcettii* and for *S. fawcettii* are similar, but they could be differentiated by Random Amplified Polymorphic DNA (RAPD) analysis (M K Tan, personal communication).

S. fawcettii var. *scabiosa* is probably a biotype of *E. fawcettii*. Even though the target pathogen (*E. fawcettii*) is present in Australia, the apparent lack of virulent strains or biotypes in Australia pathogenic to grapefruit and should qualify the Florida biotypes of *E. fawcettii* for treatment as quarantine pests. Phillips and Chandrashekar (1992) discussed the relevance of such physiological specialisation to plant quarantine.

Disease issues relating to fruit exports

Interest in the United States of America as a potential export market for Australian citrus has increased as juice prices decline and quantities of Californian navel oranges continue to be imported into Australia to compete with the Australian summer Valencia orange crop.

In 1985 the United States Department of Agriculture (USDA) completed a study of agricultural commodities entering and transiting the State of Alaska; this revealed that many commodities represented an unacceptable risk to production areas in southern States. The United States Plant Protection and Quarantine Service detected black spot (caused by *Guignardia citricarpa* Kiely) in a consignment of Australian citrus imported into Alaska and this prompted a ban on Australian citrus imports effective from 11 October 1985.

The Australian Citrus Growers' Federation (ACGF) sought clarification of the position in relation to citrus exports to the United States of America and were advised by American Embassy officials in December 1985 that quarantine barriers would remain until the incidence in Australian fruit of fruit fly, *Septoria* spot, citrus black spot and lemon scab could be clarified. ACGF approached the then Minister for Primary Industries, Mr. J. Kerrin, and expressed concern that United States fruit had free entry into Australia carrying *Septoria* sp., but Australian fruit to the United States of America was restricted entry on the basis that the *Septoria* isolates from the two countries may differ. It was agreed that collaborative research should be undertaken by scientists in the two countries to determine if variability in taxonomic parameters and pathogenicity existed in *Septoria* isolates from the two countries.

Septoria cultures isolated from citrus in Australia have often been referred to as *S. depressa* and in the United States of America as *S. citri*. Comparative studies including isozyme analyses of Australian, Californian and Arizona isolates have shown morphological variation in *Septoria* isolates from citrus in each country, but with the exception of one Queensland isolate, no substantial differences in 25 isozymes (Bonde *et al.* 1991). The results indicated that all the citrus *Septoria* isolates studied appeared to be part of a highly variable population of the one *Septoria* species, *S. citri* Pass., that occurs in the United States of America and Australia. The problem of determining pathogenic variability was not resolved.

To convince the United States of America of inland area freedom from black spot and scab, surveys of orchards in the Sunraysia-Riverland areas and in the Murrumbidgee Irrigation Areas (MIA) were carried out (Broadbent, Emmett and Wall, unpublished). These have failed to detect black spot and scab diseases or their pathogens *Guignardia citricarpa* or *S. fawcettii* var. *scabiosa*. Black spot and scab are serious diseases of coastal citrus fruit, but there are no authenticated records of these diseases or their causal fungi from inland citrus. Authorities in the

United States of America were concerned at the movement of nursery trees carrying latent black spot infections in leaves from coastal areas to inland orchards. Surveys of inland areas by Kiely in 1941 and 1947 (Kiely 1949) failed to detect *G. citricarpa* in fresh or fallen leaves, although 1 of 91 leaves from Leeton in 1942 showed copious development of spermogonia of *G. citricarpa*. Wall (1989) and Barkley (1988) could not isolate *G. citricarpa* from trees originating in coastal nurseries that had been planted in inland orchards.

Kiely's detection of *Guignardia* on citrus leaves in the MIA preceded the report of McOnie (1964) of a *Guignardia* isolated from citrus and other hosts, morphologically similar to, but physiologically and pathogenically different from the citrus black spot pathogen. It is not known which *G. citricarpa* Kiely detected as no specimens were lodged in an herbarium. As a signatory to IPPC, Australia is now obliged to maintain an effective surveillance system for diseases. Diagnostic expertise and the lodging of specimens in a national reference collection are essential to meet the Sanitary and Phytosanitary agreement under GATT.

Pest risk analysis for fruit imports

Pest risk analysis has three major stages: risk identification, risk assessment and risk management (Phillips *et al.* 1994). Options for risk management may involve sourcing fruit from regions with 'area free status' from the target pest, certification schemes and/or closely regulated postharvest sampling inspection regimes, as well as pre- and post-entry quarantine practices. This risk management depends on the Australian Quarantine and Inspection Service (AQIS) having a thorough knowledge of the industry in the area from which the fruit is sourced, including incidence of the disease, recommended control measures, their effficacy and adoption by orchards involved in export, and the postharvest treatments available and used.

Disease risks from imports of vegetative material

Protocol for citrus budwood importations

The potential dangers to Australian citrus production from introduced citrus virus and virus-like diseases are immeasurable. For example, the Asian greening pathogen and its vectors, once established in an area, have the potential to totally destroy citrus as an economic crop (Fraser *et al.* 1966; Aubert 1994). More recently, citrus variegated chlorosis, a highly destructive disease caused by *Xylella fastidiosa* (Wells *et al.* 1987) has spread through large areas of Brazil, with over two million trees rendered nonproductive (Beretta *et al.* 1993*b*).

In 1966 an embargo was placed on the importation of citrus budwood into Australia. This followed the devastation which occurred in South Africa, India and South East Asia due to greening disease (caused by a phloem-limited bacterium in the Protobacteria) and the huge tree losses experienced in California from stubborn (*Spiroplasma citri* Saglio *et al.* 1973). At that time there were no reliable indexing techniques which could be used in quarantine to detect either greening or stubborn. In the following 20 years there was no push by the Australian citrus industry to import new clones. Even now (8 years after lifting the embargo) there have been few private importations of citrus budwood. An Inter-Departmental Citrus Varieties Working Party, consisting of State Department horticulturists, initially co-ordinated departmental importations, on behalf of the Australian citrus industry, of easy-peel mandarins (e.g. Clementines from the Spanish Citrus Variety Improvement Programme), red grapefruit from improvement programs in United States of America, as well as lemons, mid-season and navel oranges. The newly formed Australian Citrus Improvement Association, representing citrus growers and nurserymen, now coordinates public importations of budwood on behalf of the Australian citrus industry.

Rigid and enforced quarantine is necessary to regulate the introduction of citrus budwood as it has the greatest potential for introducing exotic diseases. To this end a protocol for the importation of citrus budwood has been described (Roistacher *et al.* 1977; Frison and Taher 1991), and adopted by major citrus-producing countries and endorsed by FAO/IBPGR. The program involves shoot tip grafting (STG) *in vitro*, with thermotherapy, for virus elimination and full indexing of micrografted plants.

Importations of budwood into Australia have been restricted from countries with canker because of the problems inherent in detecting canker bacteria (*X. c.* pv. *citri*), particularly in tolerant cultivars. Likewise, budwood importations from countries with greening disease have also been restricted. A recent demonstration that citrus canker and greening pathogens can be eliminated by STG *in vitro* (Navarro *et al.* 1991) has resulted in the importation of budwood of two Satsuma mandarin cultivars from Japan and three orange clones from South Africa.

'Approved' or 'accredited' sources The principle that has been applied by AQIS to allow entry of temperate fruits and berries from accredited sources with no requirement for re-indexing in post-entry quarantine, is not without its dangers. 'Approved' schemes have levels of protection and consequently health status, varying from maintenance in an insect-proof screenhouse, field mother trees in foundation blocks to field multiplication or nursery trees on grower properties. The citrus experience with material from 'approved sources' demonstrates that material should not be accepted on faith as 'virus-free'. A Monroe Lisbon lemon imported as virus-free from an accredited source was found to be infected with citrus exocortis viroid (CEV) after it had been included in the New South Wales budwood multiplication planting. Its inclusion resulted in the contamination, by root grafting and mechanical transmission, of 14 trees in three lemon clones (Broadbent *et al.* 1988) and the distribution of thousands of CEV-infected buds to industry.

We are endeavouring to import citrus budwood from recognised clonal improvement programs, where horticulturally evaluated, fully indexed, STG sources are maintained in an insect-proof screenhouse. Reliability of virus-free sources overseas depends on the personnel, their dedication and skill, and the adequacy of facilities and funding. Consequently re-indexing for major citrus viruses, is undertaken on imported clones.

Strain variability Allowing entry of viruses endemic to Australia, could have disastrous consequences if more severe strains, with a wider host range, are introduced. Strains of citrus tristeza virus (CTV) are numerous and diversified (Bar-Joseph *et al.* 1981). There are new strains of CTV which can spread very rapidly and other strains which can affect many citrus rootstocks and scions once considered tolerant of CTV. It has been speculated that the stem-pitting of oranges in Peru, that has all but destroyed their orange industry, was caused by an exotic strain of CTV introduced in Satsuma orange budwood from Japan (Roistacher 1988). Like Australia, Peru already had CTV and its vector *Toxopotera citricida* (Kirk.) and had based its industry on stock/scion combinations tolerant to most strains. The recent identification of orange stem pitting strains of CTV in Queensland (Broadbent 1991) poses a threat to the entire Australian orange industry. South Australia, Western Australia and New South Wales now prohibit movement of citrus trees and budwood from Queensland. CTV strains which cause severe stem pitting in mandarins now occur in Malaysia and Thailand (Tsai *et al.* 1993).

Graft transmissible diseases of unknown cause There are a number of graft transmissible citrus diseases of unknown cause, the most important of which is citrus blight or 'declinio'. Other diseases include the gummy bark found in the Near East, North Africa and Turkey, kassala disease of grapefruit in the Sudan, the whietefly-borne citrus chlorotic dwarf virus (Korkmaz *et al.* 1994) and bud-union crease on rough lemon rootstocks. Citrus blight causes losses in Florida estimated at over 500 000 trees per year (Timmer *et al.* 1991), while 'declinio' in Brazil results in the decline of 10 million trees annually (Beretta *et al.* 1993*a*). Both are transmitted by root grafting (Timmer *et al.* 1991). Certain proteins have been associated with blight-affected trees (Derrick *et al.* 1990) and citrus blight has been detected in field trees by immunological techniques using antisera to the 12 kD blight protein (Bausher and Sweeney 1991; Derrick *et al.* 1992). The reliability of this method as an indexing tool for imported budwood has yet to be determined.

The FAO/IBPGR Technical Guidelines for

the Safe Movement of Citrus Germplasm recommend that 'whenever possible, germplasm should not be collected from high risk areas' including 'areas with diseases for which no indexing procedures exist'.

Alternate hosts Pathogenicity studies showed that strains of *X. campestris* from non-citrus hosts in Florida were capable of causing spots on Swingle citrumelo (Gabriel *et al.* 1989; Graham *et al.* 1990). These strains included strains of *X. c. fici* from *Ficus benjamina* L. and *X. campestris* from *Strelitzia reginae* Ait. Conversely, several strains of *X. c.* pv. *citrumelo* were capable of causing spots on these ornamental host plants. Strains of *X. campestris* from *Ficus* and *Strelitzia* were highly related to *X. c.* pv. *citrumelo* by DNA hybridisation analysis (Graham *et al.* 1990).

Imports of citrus relatives Alternative hosts can pose a substantial risk of introducing important citrus diseases into Australia. There was considerable consternation when centuries-old bonsais of *Murraya paniculata* (L. Jack) (gifts of the Guandong Provincial government to the New South Wales Darling Harbour scheme), were to be released from quarantine with only a fumigation treatment. *Murraya* is a citrus relative of the orange subfamily Aurantioideae, subtribe 2 Clauseninae (remote citroid fruit trees) (Swingle 1967) and a common ornamental plant here and in China. *M. paniculata* is a preferred host of the citrus psyllid (*Diaphorina citri* Kuway), a vector of greening. While showing no macro-symptoms, the bacterium-like organisms associated with greening disease have been seen in phloem cells (Aubert 1985). In addition, *M. paniculata* (*Chalcas exotica*) could be artificially infected with the canker A bacterium (Lee 1918, confirmed by Koizumi, personal communication). Elliott (1930) considered *Murraya* to be a natural host of canker.

The pathogen content of citrus relatives is not well documented. Stall and Civerolo (1991) have speculated that moderately aggressive strains of citrus bacterial spot may be pathogens of a citrus relative. A number of citrus pathogens do infect closely related citrus relatives such as *Poncirus* and *Fortunella* under cultivation, but the status of wild plants of these genera is unknown. *Microcitrus australis*, an Australian citrus relative found in northern rainforests, is a natural host of the scab fungus *S. fawcettii* var. *scabiosa* (Broadbent and Timmer, unpublished). Several citrus viruses including CTV can infect one or more citrus relatives in a number of genera including *Aegle, Aeglopsis, Afraegle, Citropsis, Eremocitrus, Microcitrus* and *Severinia* under experimental conditions. This suggests that a number of species in the subfamily Aurantiodeae of the Rutaceae may have some potential as hosts of citrus viruses (Garnsey 1988). Table 1 lists the citrus relatives in the subfamily Aurantioidiae and their susceptibility to canker, greening and citrus tristeza virus.

Preparedness for disease outbreaks

The citrus canker outbreak at Lambell's Lagoon near Darwin was detected during a routine NAQS survey. NAQS identifies potential quarantine threats from Australia's north and carries out field surveys and monitoring programs in Australia, Papua New Guinea and Indonesia. Of major concern to the small Northern Australian citrus industry are the diseases greening, canker, powdery mildew (caused by *Acrosporium tingitaninum* (C.N. Carter) Subram. and strains of citrus tristeza virus which attack mandarin, all of which are endemic to Indonesia. The possibility of canker recurring in the Torres Strait Islands owned by Australia is a real threat, given inter-island trade with Papua New Guinea. The illegal entry of boat people from China and Vietnam on the coastline of northern Australia poses a lesser risk of introducing canker or powdery mildew. Strong winds could blow greening-infected citrus psyllids (*Diaphorina citri* Kuwayama), or brown citrus aphids (*Toxoptera citricida* Kirk) infected with exotic strains of CTV, to Australian shores.

Contingency plans are necessary if an outbreak of an exotic citrus disease is to be contained swiftly. Fortunately Australia's outbreaks of canker have not been in major southern commercial citrus growing areas, but rather in remote sites or islands making successful eradication easier. It is essential that we have the mechanism in place to use personnel who are knowledgeable in the particular disease (preferably with overseas experience) for initial

Table 1 Citrus relatives in the Subfamily AURANTIOIDIAE and their susceptibility to canker, greening and tristeza

	Genus	Susceptibility to		
		Canker	Greening	Tristeza
Tribe I **CLAUSENEAE**				
Subtribe 1 Micromelinae				
I	*Micromelum*			
Subtribe 2 Clauseninae				
II	*Glycosmis*	Immune[A]		
III	*Clausena*	+[AG]	+[F]	
IV	*Murraya*	+[AG] Weakly	+[E]	+
Subtribe 3 Merrilliinae				
V	*Merrillia*			
Tribe II **CITREAE**				
Subtribe 1 Triphasiinae				
VI	*Wenzelia*			
VII	*Monanthocitrus*			
VIII	*Oxanthera*			
IX	*Merope*			
X	*Triphasia*	–[AG] Immune		
XI	*Pamburus*			+[B]
XII	*Luvunga*			
XIII	*Paramignya*	+[A]		
Subtribe 2 Citrinae				
XIV	*Severinia*	–[AG] Immune		
XV	*Pleiospermium*			
XVI	*Burkillanthus*			
XVII	*Limnocitrus*			
XVIII	*Hesperethusa*	+[AG]		
XIX	*Citropsis*	+[AG]		+[B]
XX	*Atalantia*	+[AG]		
XXI	*Fortunella*	+[AG]	+[D]	
XXII	*Eremocitrus*	+[AG]		
XXIII	*Poncirus*	+[G]	+[D]	
XXIV	*Clymenia*			
XXV	*Microcitrus*	+[AG]	+[E]	+[B]
XXVI	*Citrus*	+	+	+
Subtribe 3 Balsamocitrinae				
XXVII	*Swinglea* (*Chaetospermum*)	+[AG]		
XXIX	*Aegle*	+[G]		+[BC]
XXIX	*Afraegle*			+[BC]
XXX	*Aeglopsis*	–[G]		+[BC]
XXXI	*Balsamocitrus*	Immune[A]		
XXXII	*Feronia*	+[AG]		
XXXIII	*Feroniella*	+[AG]		

[A]Lee (1918)
[B]Muller and Garnsey (1984)
[C]McLean (1961)
[D]Aubert (1993)
[E]Aubert (1984)
[F]Van den Berg *et al.* (1992)
[G]Peltier and Frederich (1920)

diagnosis and training of survey staff in detection and hygiene. Back-up expertise in molecular biology and serology to identify and type the pathogen, overseas contacts to enable immediate dispatch of isolates for confirmation of identity (if necessary) and the quarantine facilities for pathogenicity tests are essential. The absence of an adequate quarantine containment facility has been a severe impediment to the safe and secure handling of citrus canker specimens from the recent canker outbreak in Darwin. A regulatory program should also be in place to restrict movement of trees or fruit and if necessary to defoliate and destroy trees. These plans must be worked out in consultation with the citrus growing and nursery industries. Indeed the Australian Citrus Growers' Federation has been seeking the establishment of such contingency plans for exotic citrus diseases for a number of years.

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