CITRUS GREENING DISEASE IN THE PHILIPPINES: DISTRIBUTION AND CURRENT CONTROL MEASURES

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ABSTRACT

Citrus greening disease first appeared in the Philippines in the late 1950's, probably imported with infected rootstock, and is now widespread in the Philippines. Control measures have consisted of uprooting and burning diseased trees, replanting with healthy budded plants, and periodically spraying orchards to prevent colonization by the insect vector. However, orchards originally free of both the disease and the vector have become infected, transmission of disease by the vector can be very rapid, and the vector itself can be carried large distances into healthy areas by typhoons. Recent research is concentrating on a population study of the vector and the use of natural parasites for biological control.

Citrus Greening Disease (CGD), locally known as Leaf Mottling*, was first observed in the Philippines in Batangas Province, Southern Luzon, in the late 1950's (Salibe 1966). This disease is believed to have come from China or India, since citrus seedlings had been imported into the Philippines prior to 1950 from Mainland China, Taiwan and India (Benemerito 1956). The symptoms of greening disease in Philippine citrus varieties have been described by several authors (Gonzales 1960, Salibe and Cortez 1968, Martinez and Wallace 1969, Gonzales 1983, 1987). During my visits to Taiwan in 1967, 1982 and 1985, to Thailand in 1983 and to Mainland China in 1987, I observed that the mandarin and sweet orange trees affected by this disease have similar symptoms in all these places. The findings of Lafleche and Bove (1976) that the disease was caused by a bacterium-like organism were confirmed later by Garnier *et al.* (1984) and Su (1986). The spread of greening disease to new countries is mainly due to propagation using infected planting materials, while it is naturally transmitted in local orchards by the Asian citrus psylla *Diaphorina citri* Kuwayama (Celino *et al.* 1969).

FIELD FINDINGS IN RELATION TO CONTROL MEASURES

The disease was observed to spread rapidly in the Philippines during the 1960's and early 1970's from Batangas into the adjacent Bicol region, since most of the orchards in the latter area were planted as seedlings which came from nurseries within infected areas of Batangas Province.

A field survey carried out by technicians of the Department of Agriculture Citrus Experiment Station in Lipa City in the early 1970's showed that the disease had not yet spread to other citrus producing parts of the Philippines such as Mindoro, the highlands of Northern Luzon and Mindanao. These places were categorized in the survey as healthy areas, where there was no evidence in existing plantings of the insect vector *Diaphorina citri*, and where no source of inoculum of greening disease was apparent after visual inspection. A quarantine measure through an administration order was issued by the Department of Agriculture, to prohibit the entry of infected citrus planting materials into healthy areas. In the middle of 1970's, the eradication of infected and uneconomical trees was carried out in diseased areas, where citrus growers were forced

* Also called 'likubin' in Taiwan

to change to other crops.

Indexing work conducted during the field survey in some citrus areas of the country showed that there were trees still free of the disease. However, indexing of some trees found with symptoms also showed that the disease was already present in the so-called healthy areas. It is now believed that, while sources of inoculum were present in the so-called healthy citrus areas, the insect vector was either absent, or present in such small numbers that the spread of infection was very slow. In the highland citrus areas of Northern Luzon, with a semi-temperate climate, several surveys found no trace of *Diaphorina citri* (Catling 1968).

In the later part of the 1970's, the Citrus Experiment Station established plantings of disease-free planting materials. The procedure of rehabilitation in diseased areas was to uproot and destroy infected and uneconomical trees, replant with disease-free budded plants, and periodically spray the trees to prevent colonization by the insect vector. At the same time, the citrus station technicians encouraged nurserymen to establish nurseries only in isolated locations, to select propagating materials from mother trees existing in healthy areas, and to examine all planting materials carefully to make sure they were free of disease symptoms, while all the time preventing infestation by citrus psylla of nursery seedlings, rootstock and budded plants. The orchards of the Citrus Experiment Station in Lipa City served as a demonstration area, and showed the success of the rehabilitation program in areas severely infested with the disease (Gonzales 1981). Since then, the experimental field of the citrus station has been used for studies on monitoring the insect vector, population dynamics of the vector, modes of reinfection with the disease, and the economics of periodic chemical control of the insect vector.

The technology generated on the control of greening disease at the Citrus Experiment Station within the diseased area has encouraged farmers in nearby inland and island provinces which are considered healthy areas to plant citrus orchards. The demand of citrus growers for planting materials has run up against the existing quarantine regulations. In many instances, not only are nurserymen in infected areas tempted to sell their budded plants to outside growers, but the growers themselves in healthy areas, in their desire to plant citrus, have connived by smuggling in seedlings. It is under these conditions that large areas have been planted in citrus. It is estimated that since 1981, growers in the provinces of Batangas, Oriental Mindoro, Laguna and Rizal have planted about three million citrus trees, while two million more trees have been planted in orchards scattered through other provinces of Luzon. About 80% of these planting consist of Szinkom mandarin, and 20% of calamondin. In the citrus growing areas of Davao Province in Mindanao, however, citrus growers concentrate mainly on pummelos and King mandarin, while in the temperate highlands of Northern Luzon the most common varieties being planted are various kinds of sweet oranges. Altogether, the area of citrus orchards in the Philippines increased from 25,150 ha in 1981 to 33,800 ha in 1987.

CURRENT PROBLEMS IN DISEASE CONTROL

Control of the Insect Vector

One major problem is the difficulty of controlling the insect vector. This is reflected in the following field observation.

- The orchards at the Citrus Experiment Station were re-planted with disease-free plant materials in 1976-1980, and since then have been closely monitored for the presence of the vector at either the nymphal or the adult stage. They have also been subjected to a regular schedule of spraying with chemical pesticides. There have been instances when even though no insect vectors were observed before isolated orchards were sprayed with strong insecticides, they were recorded as present just a few days later. In some cases, isolated citrus orchards which were originally free of both the disease and the vector have been observed with disease symptoms.
- Foundation plantings of citrus surrounded by sugarcane plantations, where there were no other citrus plantings within a radius of 5 km or more, have been infested with adults and nymphs of the insect vector after a strong typhoon.
- In a very large, isolated orchard of disease-free citrus trees of bearing age in east Davao, Mindanao, there was an upsurge of large numbers of citrus psylla. These were immediately eradicated by chemical spraying. Thereafter, the orchard was monitored weekly for insect vectors, which were always controlled at zero level. A year later, about 8,800 trees had to be

uprooted due to severe infection with greening disease (Catling 1985).

At the Baguio Experiment Station, located in semi-temperate highlands in Northern Luzon, all existing citrus plants were removed prior to the introduction and planting of imported rootstock and scions from U.S.A., all of which were certified free of any disease. The area around the station was surveyed many times and found to be free of the insect vector. During the recent (April 1988) visit of the FAO Regional Project Coordinator Dr. B. Aubert, six adults of citrus psylla were found in an orchard containing all imported citrus scion-stock combinations, and some trees are already manifesting symptoms of greening disease.

Spread of Disease without the Presence of the Insect Vector

In Laguna, Southern Luzon, greening disease appears to be spreading from tree to tree although so far no insect vector has been found, after many visits by citrus technicians. The same situation is also found on the Eastern side of the island of Mindoro (Mindoro Oriental) and in some citrus orchards in Davao City, Mindanao.

Complex Infection with Other Virus Diseases

Different varieties of citrus vary in their response to greening disease (Salibe and Cortez 1968, Gonzales and Viñas 1981). Research has shown that pummelos, calamondin, lime and lemon are more resistant than other species and varieties to infection with greening disease. Lately, it has been observed that pummelo trees in some orchards in Davao area are declining and becoming unproductive. The effect of severe tristeza infection on the trees, which also harbor the greening pathogen, is now being studied.

NEW CONTROL PROGRAM FOR GREENING DISEASE

A four-year control program was begun in 1988, with the assistance of the Food and Agriculture Organization (FAO) and the Food and Fertilizer Technology Center for the Asian and the Pacific region (FFTC/ASPAC). This program was built in to the existing Philippine Citrus Development Action Program (PCDAP).

Intensive Study of the Insect Vector

Colored sticky traps, consisting of Saturn yellow scothal adhesive and tanglefoot, are being used to monitor the *Diaphorina citri* population, in order to establish the distribution of the species and determine its population at different altitudes and latitudes and under different climatic conditions, in both forest and farmland (Samways 1987, Aubert and Ouilici 1988). This will make clear some aspects of the biology of *D. citri*, such as its temperature preferences and the effect of low temperature on its survival. These traps are now installed at different locations in Luzon and Mindanao. Traps in the midst of citrus production areas will also show the influence of the insect breeding cycles on population density. The presence of other major tropical pests of citrus can also be determined by the traps, and will supply information as a basis for practical pest management all year round.

The Use of Parasites for Biological Control

A method has been developed for discovering local natural enemies or parasites of the insect vector. This method can determine if parasitic wasps exist which attack the citrus psylla nymph, or if there are efficient chalcidoid parasites. This will provide more accurate knowledge of the existing ecological equilibrium, and will enable us to devise strategies of biological control and implement integrated citrus pest management. The introduction of exotic natural enemies will also be implemented. The parasite *Tetrastichus radiatus* will be raised and reproduced in the specially designed rearing laboratory under quarantine at the Citrus Experiment Station in Lipa City, Luzon area and in Davao City, Mindanao area. The methodology developed in Reunion Island and Taiwan for the successful establishment of this active ectoparasite will be adopted (Aubert 1987, Chiu and Chien 1987). In view of the success of the biological control programs using this parasite in Reunion and Taiwan, we can expect a similar sharp reduction in the number and severity of outbreaks of the insect vector *Diaphorina citri* in citrus areas after the widespread release of this parasite.

Establishment of an Epidemiological Data Base

In order to carry out integrated citrus pest management in infected areas, epidemiological indicators of greening disease need to be established. To achieve a realistic epidemiological data base, the severity of disease will be rated according to the method used for the epidemiological interpretation of disease incidence in Reunion Island (Gottwald and Aubert 1987). The methodology of periodically giving canopies of either young trees or trees of bearing age in given plots a rating according to disease severity will provide adequate points for modeling within three years. Models of disease spread will be correlated with those of the insect vector and those of any parasite population over time, so that both spatial and temporal aspects of disease progress can be analyzed.

An example of the modeling of natural transmission of greening disease in Batangas Province is the identification of two new citrus orchards two to three years old in which only a few plants were infected. These orchards were adjacent to a grove of five to seven year old trees which showed infection with greening disease. Both the disease and the insect vector were present in all three orchards. Disease spread in this case will hopefully allow us to model increase in a situation where disease was introduced from contaminated planting stock. Incidentally, none of the three orchards have been sprayed with insecticides since they were planted, and therefore the vector populations should be high and fluctuate naturally.

Models resulting from several sets of trials now being conducted in Luzon and Mindanao will be compared to restrained and unrestrained disease models by a statistical comparison of the progress curve (AUDPC) of the disease in the contaminated areas, by correlation, or other methods. We hope that this will allow us to develop useful and practical methods of identifying better disease control strategies for each location.

ETIOLOGY

With the support of FAO, collaborative work will be carried out with the BCM laboratory (INRA, Bordeaux, France) or with the laboratory network organized in Asia by FFTC/ASPAC, in order to culture the citrus greening organism and produce monoclonal antibodies. Samples of the following plant materials are to be sent to laboratories abroad.

- Young orange or mandarin seedlings inoculated with *D. citri*, harboring greening disease only. Inoculated plants showing greening disease infection will be sent to the Laboratory with its roots in peat moss.
- Consignments of samples of 30 leaves from mature trees showing typical symptoms of leaf mottling, twig dieback, lopsided fruit and aborted seeds. Leaves showing infection with citrus canker or attacked by insect pests will be discarded.
- Periwinkle plants (*Catharanthus roseus*) inoculated with the greening organism by dodder (also known as love vine: *Cuscuta campestris*), bridged with infected citrus. The entire periwinkle plant will be sent with its roots in peat moss.

The above samples will help in testing for polyclonal and monoclonal antibodies. The scientists at INRA-Bordeaux BCM, France, have reported recently that with such collaboration, enough monoclonal antibodies should soon be available for the development of serological techniques to detect citrus greening in citrus nurseries and orchards (Garnier *et al.* 1987).

PRODUCTION OF DISEASE-FREE CITRUS PLANTING MATERIALS

A program of indexing and selecting candidate citrus parent trees for budwood certification is now being pursued, with the establishment of disease-free foundation plants as a source of propagating material. The process includes obtaining clean planting materials either from cooperating citrus centers abroad, or by asking foreign laboratories to clean budwood samples sent to them of the existing citrus varieties in the Philippines. The Citrus Experiment Station in Lipa City has also established a facility for using a modified technique of citrus shoot-tip grafting and rapid propagation methods, to obtain citrus budwood free of greening disease and other citrus viruses.

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DISCUSSION

Q. (M. Koizumi)
You mentioned that disease-free trees were planted in safe highland areas, and that no vectors were observed to be present, but that it is still possible to find some diseased trees. How did transmission of the disease occur?

A. The probable situation is that there was some introduction of infected propagating materials. It is also very easy for the vector to colonize new areas, and for several years we neglected to monitor highland orchards for the vector. It is possible that some colonization occurred, but I think infection from diseased planting materials is more likely.

Comment: (B. Aubert)
I should like to make some comments on the population dynamics of *Diaphorina citri*. Many factors are not yet understood, and basic studies should be continued on this vector. Weather has a strong influence. In areas of high rainfall and high relative humidity, the population levels of *D. citri* will be relatively low. Populations of *D. citri* reach a peak in dry seasons or fairly dry climates. Since there is then high fertility, populations of *D. citri* grow very fast. In the highlands you mentioned, the insect vector may be present as an endemic, but the entomologist may not be able to find it in humid weather when it is present in very small numbers and conclude that it isn't there. However, he might come back two months later, during a short dry period, and find populations of the vector.

Another point you mentioned was the natural parasites of the vector. We have some interesting information on this point from Reunion Island, which has a very varied climate ranging from very dry to very humid, which offers a great deal of information on biological control.