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Transmission of Indian Citrus Decline by *Trioza erytreae* (Del Guercio), the Vector of South African Greening

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Two psyllid vectors of greening are known. In Africa the disease agent is transmitted by *Trioza erytreae* (Del Guercio) (McClellan and Oberholzer, 1965), while in India, China, and south-east Asia the vector is the oriental psylla, *Diaphorina citri* (Kuwayama) (Capoor *et al.*, 1967).

Two forms of greening have been characterized (Bové *et al.*, 1974). One is heat sensitive (no symptoms above 27 to 32°C); it is the form present in South Africa. The other form is heat tolerant (good symptoms at 30 to 35°C) and is prevalent in India (citrus decline), Taiwan (likubin), the Philippines (leaf mottling), and China (yellow shoot). The Asian, heat-tolerant form of the disease is much more destructive than the heat-sensitive, African form.

MATERIALS AND METHODS

The Indian greening used in this work was from Poona and has been described earlier (Bové *et al.*, 1974); it is free of any other known pathogen. Young adults of the African citrus psylla, *T. erytreae*, collected near Pretoria on citrus trees showing no symptoms of greening were carried to Bordeaux, France by Dr. D. Catling. On December 30, 1971 they were placed in insect-proof cages on healthy young Hamlin sweet orange seedlings. These cages were kept in a large growth chamber held at $24 \pm 1^\circ\text{C}$ during the 16-hour light period (15,000 lux) and at $21 \pm 1^\circ\text{C}$ in the dark (8 hours). Relative humidity was 80 ± 10 per cent.

After one month a new generation of psyllas was obtained and the young adults were again transferred to healthy seedlings under the same conditions. This

Even though Koch's postulates have not yet been fulfilled, it is probably that the procaryote-like organism associated with all forms of greening and their psyllid vectors in the casual agent of the disease (Bové and Saglio, 1974).

Since South Africa seems to be infested only with the less destructive, heat-sensitive form of the greening pathogen the introduction of the more virulent, Asian form would be detrimental to the South African citrus industry, especially if the African vector, *T. erytreae*, the only one known to be present in South Africa, should prove capable of transmitting the disease. The work reported here shows that *T. erytreae* can indeed transmit the destructive, heat-tolerant Indian greening.

procedure was repeated twice. Three psylla generations were thus obtained. The adults from the third generation were used for acquisition feeding on a Hamlin sweet orange seedling with Indian (Poona) greening. This seedling showed severe symptoms of the disease but still had young flushes of growth. The adult psyllas from the third generation were left on the diseased Hamlin seedling until eggs had been laid and were then removed and discarded. About 100 adults (fourth generation) were produced; they were divided among 5 healthy sweet orange seedlings and each seedling was put in an individual cage. Transmission feeding was from April 20 to May 9, 1972.

Electron microscopy techniques are described elsewhere (Garnier *et al.*, 1976).

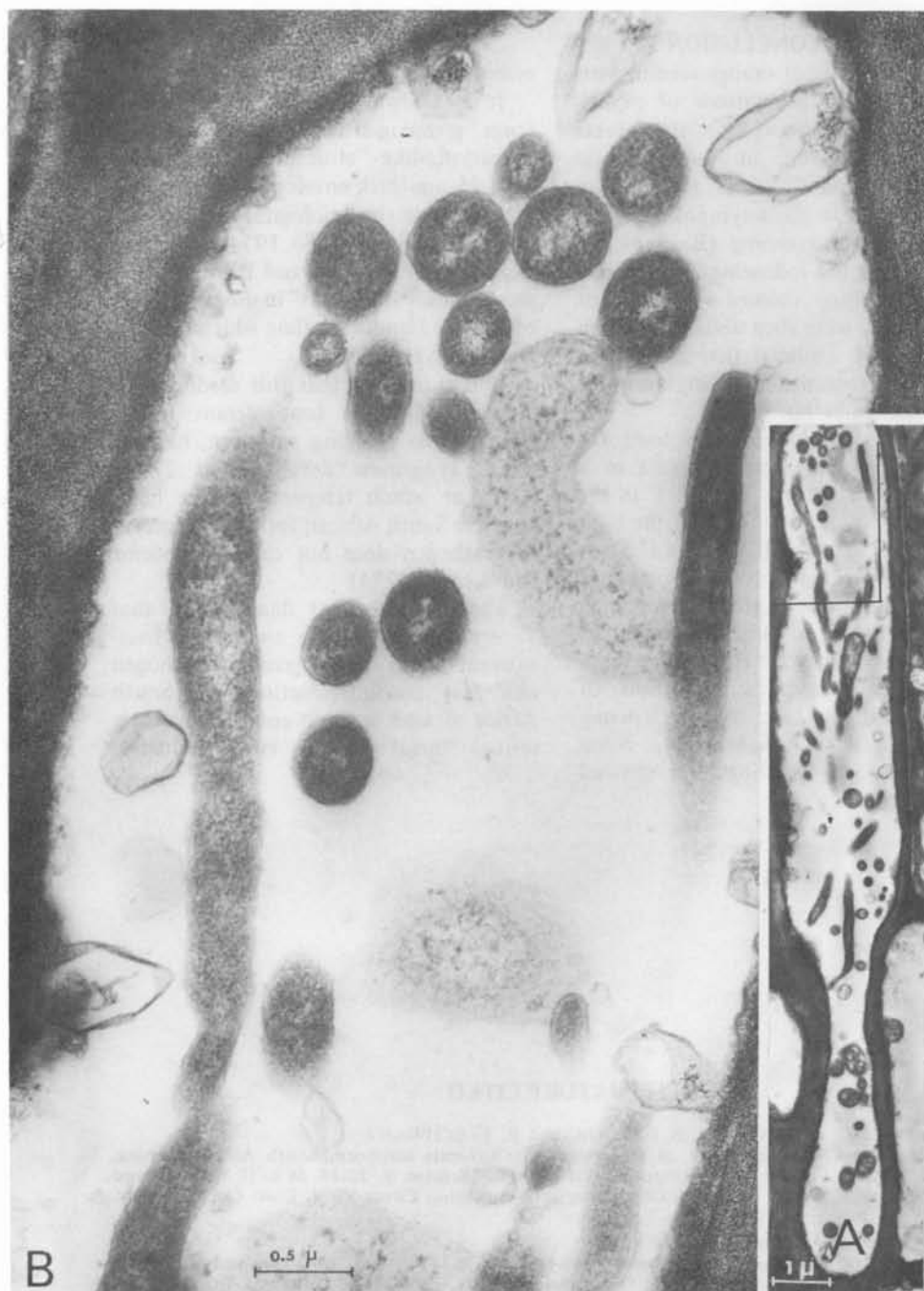


Fig. 1. (A) Electron micrograph of a thin section through the phloem of a Hamlin sweet orange seedling showing symptoms of greening 10 months after transmission feeding by *Trioza erytrae*; bodies present in the sieve tube represent the procaryote-like structures associated with greening and are probably the etiological agent; (B) enlargement of the upper part of A.

RESULTS AND CONCLUSION

The Hamlin sweet orange seedlings on which the three generations of psyllas were reared were kept, after the insects had been removed, in a greenhouse between 20 and 27°C, a temperature range adequate to allow symptom expression of African greening (Bové *et al.*, 1974). During the following 2 years none of these seedlings showed symptoms of greening; they were then discarded. It can thus be safely assumed that the psyllas used for the acquisition feeding were free of the greening pathogen.

The five Hamlin seedlings used for transmission feeding were placed in a growth chamber kept at 27 ± 2°C in the dark (8 hours) and 32 ± 2°C in the light (16 hours at 20,000 lux). This temperature range favors symptom expression of heat-tolerant strains of greening to which the Poona, Indian form belongs. After 10 months, one of the five Hamlin seedlings began to show typical symptoms of greening: leaf mottling, zinc deficiency patterns, young twig dieback, etc. None of the other four seedlings ever showed

symptoms.

It is known that the phloem tissue from greening-affected citrus contains procaryote-like "structures," surrounded by a 25 nm-thick envelope system, which probably are the etiological agent of the disease (Bové and Saggio, 1974; Garnier *et al.*, 1976). Figure 1A and B (next page) shows these "structures" in the sieve tubes of the one Hamlin seedling which showed greening symptoms.

It is thus clear that this seedling was infected with the heat-tolerant Indian form of the greening pathogen, because severe symptoms developed at 27 to 32°C, at which temperatures the heat-sensitive South African form of the greening pathogen does not cause symptoms (Bové *et al.*, 1974).

These experiments demonstrate that *T. erytrae* is able to transmit a heat-tolerant strain of the greening pathogen and that the introduction into South Africa of such a strain could represent a serious threat to the citrus industry.

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