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# Survey of the Incidence of Citrus Greening Disease and its Psylla Vector in Nepal and Bhutan

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**ABSTRACT.** Following the inception of the Citrus Greening Disease Investigation Scheme in 1972, a survey of the disease and its psylla vector, *Diaphorina citri*, has been carried out in 28 districts of Nepal and one district of Bhutan using field symptoms, presence of the gentisoyl glucoside marker and presence of the Greening bacterium in tissue for diagnosis. Both the disease and vector were recorded in all districts, the most reliable indicators being symptoms and presence of the marker. Electron microscopy of infected tissue detected the organism in samples from only four districts. The positive effects of biological control of the psylla using two species of parasitic wasp were observed in some districts.

Citrus greening disease was first reported in Nepal in 1967 where it was observed in the Pokhara valley (5,6). It was presumed to have been introduced in infected plant material from

India. Thousands of trees have been killed in Nepal, and it is spreading to other areas of the country (6). The psylla vector, *Diaphorina citri*, is found in virtually all the citrus growing areas (8), and it has played an important role in the spread of the disease (9,10).

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TABLE 1  
RESULTS OF SURVEYS OF CITRUS GREENING DISEASE IN NEPAL AND BHUTAN

District	Field symptoms	Marker substance	GO detected by EM	Psylla vector	Parasitic wasp
Nepal					
Illam	**	*	— <sup>z</sup>	*	—
Dhankuta	*	*	—	*	—
Solukhumbu	*	*	—	*	—
Udaipur	*	*	—	*	—
Ramechhap	*	*	*	*	—
Sindhuli	*	*	*	*	*
Dhanusa	*	*	*	*	*
Nawalparasi	*	—	—	*	—
Chitwan	*	*	—	*	—
Makawanpur	*	*	—	*	—
Kavre	*	*	—	*	—
Kathmandu	*	*	—	*	*
Dhading	*	*	—	*	—
Gorkha	*	*	—	*	—
Lumjung	*	*	—	*	—
Kaski (Pokhara)	*	*	*	*	*
Tanhun	*	*	—	*	—
Syangja	*	*	—	*	—
Parbat	*	*	—	*	—
Baglung	*	*	—	*	—
Myagdi	*	*	—	*	—
Palpa	*	*	—	*	—
Gulami	*	*	—	*	—
Pyuthan	*	*	—	*	—
Dang	*	*	—	*	—
Salyan	*	*	—	*	—
Dadeldhura	*	*	—	*	—
Baitadi	*	*	—	*	—
Bhutan					
Phuntsholing	*	*	—	*	*

<sup>z</sup> \* = present — = absent

Citrus greening is caused by a phloem-limited bacterium which can be detected by electron microscopy (4). The disease can be detected by thin layer chromatography (TCL) to show the presence of the fluorescent marker, gentisoyl glucoside (7).

A regular survey has been conducted since 1972 in 28 districts of Nepal and one district of Bhutan. Diagnosis was based on field symptoms, TLC of bark extracts, and electron microscopy of phloem tissue in leaf and fruit columellas. Samples for electron microscopy were done in Japan by Dr. J. Imada. The presence of the vector and its parasitoid wasps was also noted, since biological control using *Tamaraxia radiata* has been effective

in Reunion and parts of Nepal (1,8). Table 1 shows the results of the survey. Greening symptoms were recorded in all districts, and the marker substance was detected in all but one Nepalese district. The greening bacterium, however, was detected, by electron microscopy in samples from only four districts.

*Diaphorina citri* was also noted in all districts, while the primary parasites, *T. radiata* and *Diaphorencyrtus aligarhensis* samples of which were sent to Reunion for identification, were observed in five districts. In the Phuntsholing district of Bhutan, 90% of the psylla nymphs were parasitised. In Pokhara the mild parasite *Allographa* sp. was also recorded.

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