High incidence of *Sugarcane yellow leaf virus* (SCYLV) in sugar plantations and germplasm collections in Thailand

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Abstract. Sugarcane plantations and germplasm collections from across Thailand were tested in two surveys within the years 2000–2003 for *Sugarcane yellow leaf virus* infection. Twenty-five to 100% of cultivars tested at each plantation/ germplasm collection were infected, among them those which had been imported from international breeding stations. Plantation management based on resistant cultivars or virus-free seed cane plantation practices is proposed.

The new sugarcane disease Yellow Leaf Syndrome (YLS) was described in the 1990s in Hawaii (Schenck 1990), Brazil (Vega 1994), Florida (Comstock *et al.* 1994) and Africa (Bailey *et al.* 1996). The luteovirus *Sugarcane yellow leaf virus* (SCYLV) was identified as causal agent of the disease (Scagliusi and Lockhart 2000). Further tests for SCYLV by tissue blot immunoassays and/or PCR (Schenck *et al.* 1997; Korimbocus *et al.* 2002) revealed that SCYLV also occurred worldwide. The worldwide distribution most likely proceeded through germplasm exchange and it depended very much on whether the imported germplasm was susceptible to and infected by SCYLV. Spread of SCYLV usually occurs by vegetative propagation of infected stem pieces (so-called seed pieces). It can also be facilitated through the vector activity of viruliferous black sugarcane aphid

(*Melanaphis sacchari*), but infection rates are generally slow (Lehrer *et al.* 2007). SCYLV-infected plants may be free of obvious symptoms (Lehrer and Komor 2008); thus infections and subsequent spread of SCYLV may fail to be noted, resulting in underestimation of the problem. Sugarcane is an important commodity for Thailand and disease control of sugarcane is, therefore, of vital economic interest. This report shows that SCYLV is widespread in Thai plantations and germplasm collections, and that SCYLV-free management of cane fields for seed pieces and of plantation fields is recommended.

The surveys were conducted from October to February in the years 2000–01 and 2002–03. Samples were taken from the uppermost, fully expanded leaf from 6–12-month-old sugarcane plants (*Saccharum* spp. hybrids). Leaf pieces of ~15 cm length

 Table 1.
 SCYLV in samples collected from plantations, germplasm collections and cultivar collections of sugar mills in different

 Thai sugar-producing areas

Samples were collected from commercial fields, from cultivar collections of sugar mills and from test fields in research and breeding stations. The tested cultivars in the sugar mill collections were those which were already known to exhibit YLS

| Name (place, province, region) | Infected cultivars/ total tested 2000-01 | Infected cultivars/ total tested 2002-03 |
|--|---|---|
| Plantation fields | | |
| Satien Farm (Kanchanaburi, Central Thailand) | 3/12 | 3/13 |
| Fields in Chonburi (Chonburi, Eastern Coast Thailand) | 13/21 | |
| Suparb Farm (Udon Thani, North East Thailand) | 0/13 | |
| Nong Khum (Prachinburi, Central Thailand) | | 0/5 |
| Research and quarantine centers | | |
| Ta Maung Sugarcane Center (Kanchanaburi, Central Thailand) | 24/84 | |
| Supanburi Field Crop Research Center (Supanburi, Central Thailand) | 28/97 | |
| Mitr Phol Quarantine Phu Kieo (Chaiyaphum, North East Thailand) | 25/112 | |
| Sugar mills (cultivars expressing YLS symptoms) | | |
| Pranburi Sugar Mill (Pranburi, Prachuapkhirikhan, Southern Thailand) | | 3/4 |
| Buriram Sugar Mill (Buriram, Buriram, Southern North East Thailand) | | 6/6 |
| Kumphawapi Sugar Mill (Kumphawapi, Udon Thani, North East Thailand) | | 13/13 |
| Mitr Phol Sugar Mill (Chayapum, Chayapum, North East Thailand) | | 12/12 |

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Fig. 1. Distribution of SCYLV-infected sugarcane in different sugarcane-growing regions of Thailand. The circles correspond to the location where the samples for the SCYLV-surveys were collected. The shaded segments of the circles represent the proportion of cultivars testing positive for SCYLV by tissue blot immunoassay.

were collected in a plastic bag with damp tissue enclosed. Within 5 h of sampling, the blade was stripped from the midrib and a freshly cut cross-section of the midrib was pressed onto a nitrocellulose membrane (Biorad TransBlot membrane, $0.2 \,\mu\text{m}$ pore size). Three prints were made from each midrib, each from a freshly cut surface. This tissue blot immunoassay occasionally failed to detect SCYLV infections in extremely chlorotic leaf samples (data not shown) and in these instances tissue prints from root sections were used. The membranes with the prints were stored until subsequent processing (Fitch *et al.* 2001). The leaf sample was considered as infected by SCYLV when at least one bundle of the cross-section showed colour deposits in each of the three prints.

Leaf samples from more than 300 sugarcane cultivars from plantations and germplasm collections in the main sugarcane areas of Thailand were tested for SCYLV in the years 2000–01 and 2002–03. SCYLV was found in all sugarcane regions and on average 27% of the tested cultivars were infected. (Table 1). A second survey (2002–03) collected samples from cultivars which expressed YLS-like symptoms on a regular basis in the fields of sugar mills. Nearly all of these cultivars showed SCYLV-infection (Table 1). Many of the tested cultivars had been imported from breeding stations outside Thailand. Imports from Fiji exhibited a lower proportion of infected cultivars (17%) than cultivars from Canal Point, FL, USA and Kantalai, Sri Lanka (84–100%).

A positive immunoreaction of the tissue blot immunoassay confirms the presence of the virus. However, due to low virus titre, particularly in older leaves, some infections may fall below the sensitivity threshold of the immunoassay (Lehrer and Komor 2008). Here we confirm that positive detection of SCYLV by tissue blot immunoassay depended on the leaf age. Whereas all samples from young source leaves (#1 to #3) showed infection, only one to two-thirds of the tissue prints from older leaves were positive, although the SCYLV-infection was most likely present in the phloem of all leaves and some of these leaves even expressed YLS-symptoms (Table 2).

The survey showed that plantations and breeding stations in all sugarcane areas of Thailand are infected by SCYLV (Fig. 1). Although YLS does not destroy the entire harvest, it reduces yield

Table 2. Detection of SCYLV by tissue blot immunoassay in leaves of differing age sampled from two stools exhibiting YLS-symptoms The plant was growing in a plantation field in Sra-Kaew, Buriram. The uppermost, fully expanded leaf was numbered #1 (top-visible dew-lap leaf), with subsequent leaves numbered downward consecutively. Each stool had 8–9 stalks (including the side stalks). Each sample represents an

individual leaf

| Leaf | Symptom | Positive samples/ total |
|------|----------------|----------------------------|
| #1 | No symptoms | 3/3 |
| #2 | Faintly yellow | 8/8 |
| #3 | YLS | 8/8 |
| #4 | YLS | 3/8 |
| #5 | YLS | 11/17 |
| #6 | YLS | 3/9 |
| #7 | Pale yellow | 6/9 |
| #8 | Yellow line | 2/5 |

up to 30%, even when the plants are asymptomatic (Lehrer *et al.* 2001). Yields are further decreased when plants are infected by SCYLV in combination with phytoplasma (Aljanabi *et al.* 2001). Two combined strategies are proposed to confine SCYLV-infection to a low level. One is to identify and deploy resistant varieties (Schenck and Lehrer 2000). UT 91-2-633 appears to be a Thai resistance candidate according to our survey. No infection was noted in eight samples of this cultivar, but further work would be required to confirm this observation (0/8 samples from one site only).

The other strategy is a cultivation scheme in which virus-free cane plants, generated by meristem tip culture, are grown for seed piece production in fields remote from commercial sugarcane fields. It has been shown that a sugarcane-free gap of a few hundred metres, or a boundary of resistant cane of a similar distance, was sufficient to prevent *de novo* infection by aphids (Lehrer *et al.* 2007). The necessary distance needed for the plantations in Thailand would have to be determined. Whether SCYLV is spreading further in Thailand or has already reached a steady-state of infection would have to be determined by further surveys.

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