Long-term association of tomato yellow leaf curl virus with its whitefly vector *Bemisia tabaci*: effect on the insect transmission capacity, longevity and fecundity

Galina Rubinstein and Henryk Czosnek

Department of Field Crops and Genetics, and the Otto Warburg Centre for Biotechnology in Agriculture, Faculty of Agriculture, The Hebrew University of Jerusalem, Rehovot 76100, PO Box 12, Israel

The association between tomato yellow leaf curl geminivirus (TYLCV, Israeli isolate) and its insect vector, the whitefly *Bemisia tabaci*, was investigated. Insects that emerged during a 24 h period were caged with TYLCV-infected plants for a 48 h acquisition access period, then with egg-plants – a TYLCV non-host – for the rest of their lives. While TYLCV DNA was associated with the whiteflies during their entire adult life, the amount of capsid protein rapidly decreased and was not detectable in the insect after approximately 12 days of age. The ability of the infected whiteflies to transmit TYLCV to tomato test plants steadily decreased with age but did not disappear completely. Transmission by viruliferous insects decreased from 100% to 10–20% during their adult lifetime, compared with a decrease from 100% to 50% for non-viruliferous insects. The association of TYLCV with adult *B. tabaci* led to a reduction of 17–23% in their life expectancy compared with insects that had not acquired the virus, and to a 40–50% decrease in the mean number of eggs laid. These results suggest that TYLCV has some features reminiscent of an insect pathogen.

Introduction

Tomato yellow leaf curl virus (TYLCV) is a group of whitefly-transmitted geminiviruses (Cohen & Harpaz, 1964; Czosnek et al., 1988) that causes extensive damage to tomato crops in many tropical and subtropical regions worldwide (Czosnek & Laterrot, 1997). The genome of TYLCV is either monopartite (Mediterranean isolates) or bipartite (Thailand isolate) (reviewed by Rybicki, 1994; Padidam et al., 1995). While the acquisition and transmission of TYLCV by whiteflies have been studied in some detail (Cohen & Nitzany, 1966; Zeidan & Czosnek, 1991; Mehta et al., 1994; Caciagli et al., 1995), the interactions between this geminivirus (as well as others) and *Bemisia tabaci* are still poorly understood. In a manner similar to that for other whitefly-transmitted geminiviruses (Duffus, 1987), *B. tabaci* transmits TYLCV in a persistent, circulative manner (Cohen & Nitzany, 1966). The virus requires a latent period of 12–24 h in the vector (Cohen & Nitzany, 1966). The genomic DNA of TYLCV can be detected in *B. tabaci* individuals 30 min after the beginning of acquisition feeding, and it accumulates in the insects as feeding proceeds (Zeidan & Czosnek, 1991). The virus nucleic acid remains associated with the insects for many days after they have ceased to feed on infected tomato plants (Zeidan & Czosnek, 1991; Cohen & Antignus, 1994). Earlier data suggest that at least some geminiviruses are reminiscent of insect pathogens and are deleterious to their whitefly vector. In earlier investigations it was shown that the presence of TYLCV in whiteflies was accompanied by the induction of antiviral factors (Cohen, 1967, 1969). Moreover, it was reported in an abstract that squash leaf curl geminivirus (SqLCV) invaded a number of whitefly organs, tissues and cells and was associated with gross as well as ultrastructural abnormalities of the reproductive, digestive and excretory systems (Pesic-van Esbroeck et al., 1995). In this paper, we present evidence that long-term association of TYLCV with its whitefly vector affects the transmission capacity, longevity and fecundity of the insect.

Methods

- Maintenance of virus cultures, whiteflies and plants. Cultures of an Israeli isolate of TYLCV (Navot et al., 1991) were maintained in tomato plants (*Lycopersicon esculentum* cv. FA144) and propagated by whitefly-mediated transmission. *Bemisia tabaci* of the B biotype (Cohen,
Acquisition and transmission of TYLCV by B. tabaci. Adult whiteflies were caged with leaf number 2 (true leaf-numbered from apex downwards) of TYLCV-infected tomato plants (at the six-leaf stage, approximately 3 weeks after inoculation) for a 48 h acquisition access period. The insects were then collected and reared on egg-plants (Solanum melongena cv. Classic), a TYLCV non-host. Groups of three viruliferous insects (minimum number of insects to ensure 100% transmission) were caged with leaf number 2 of each tomato test plant (at the four-leaf stage) for a 48 h inoculation access period. The plants were then sprayed with insecticide and grown in an insect-proof growth chamber. Infection was appraised 4 weeks later by hybridizing squashes of the plants’ youngest true leaf (Navot et al., 1989) with radiolabelled plasmid pTYH19 containing a full-length copy of the TYLCV genome (Navot et al., 1991) and autoradiographing for 24 h, and by appearance of symptoms.

Analysis of TYLCV DNA and capsid protein associated with whiteflies. DNA from groups of 10 insects was prepared as previously described (Zeidan & Czosnek, 1991). One-tenth of the DNA was subjected to electrophoresis, blotted and hybridized with radiolabelled plasmid pTYH19. Protein extracts were prepared from groups of 10 insects as described by Kunik et al. (1994). One-fifth was submitted to SDS–PAGE (Laemmli, 1970) and immunoblotted with an antiserum against a purified preparation of TYLCV virions (a gift from B. Gronenborn) as described by Kunik et al. (1994). Reactive protein bands were visualized using the enhanced chemiluminescence procedure (ECL Western blotting, Amersham). DNA signals were quantified using the Personal Densitometer (Molecular Dynamics).

Measurement of insect mortality. Whiteflies were reared in a nethouse with temperatures close to those in the open. Adult insects that emerged within a 24 h period were caged with infected tomato plants. Control insects were caged with non-infected plants. The insects were collected after 48 h and raised on egg-plants, a TYLCV non-host. For each plant (15 plants for each insect population), 10 insects were caged with leaf number 3 of plants at their four-leaf stage. The insects were collected and placed on new egg-plants every 2 weeks to avoid emergence of new adults. The number of insects (living versus dead) was counted every 3–4 days.

Measurement of insect fecundity. Adult insects of various ages after emergence were caged with TYLCV-infected tomato plants for 48 h. Control insects were caged with non-infected plants. The insects were then caged with egg-plants (one insect per plant) using leaf cages. The number of eggs laid was counted after various periods of time. The number of emerging instars was also counted.

Statistical analyses. The effect of TYLCV on insect mortality and on insect fecundity was analysed by the $\chi^2$ test and by the Student’s $t$ test, respectively, using the JMP statistical analysis package version 3.1 (SAS Institute, 1995).

Results

Long-term association of TYLCV with B. tabaci: retention of viral DNA, capsid protein and ability to transmit the virus

Adult whiteflies that emerged during a 24 h time period were caged with TYLCV-infected tomato plants. After a 48 h acquisition access period, the 3-day-old viruliferous insects were reared on egg-plants for the rest of their lives. Every 3–5 days (until all insects had died of old age) groups of 70 insects were collected randomly. Fifty insects were used to test virus transmission (three insects per plants, 15 plants per time-point), 10 were used for analysis of viral DNA and 10 for viral capsid protein. As shown before, viruliferous whiteflies contained a single detectable viral DNA molecule (the viral genomic DNA), appearing as a single band in Southern blot hybridization of individual insects (Zeidan & Czosnek, 1991). The viral capsid protein appeared as a single ~ 30 kDa protein band in Western blots (Kunik et al., 1994) and was best detectable when pooled protein extracts, equivalent of two insects, were used. Plant infection was tested 4 weeks after inoculation.

Fig. 1 shows that detectable amounts of viral DNA were associated with the insects during their entire life. In parallel, the amounts of viral capsid protein associated with these insects rapidly decreased and ceased to be detectable approximately 12 days after TYLCV acquisition. The ability of the insects to transmit the virus to tomato test plants steadily decreased during the lifetime of the insects (from 100% to 10–20% transmission), but did not entirely disappear (all insects died 33 days after virus acquisition).

The question of whether ageing alone accounted for the decrease in virus transmission was investigated by comparing transmission efficiency of insects that acquired virus at various ages with that of insects that acquired virus immediately after emergence. Insects collected during a 24 h period after emergence were reared on egg-plants. After various time periods, groups of insects were caged with infected tomato plants for a 48 h acquisition access period, then with tomato test plants for a 24 h inoculation period, as described above.
The ability of these insects to transmit the virus to test plants was compared with that of insects that have acquired the virus immediately after emergence (as described above). The tomato plants were analysed for the presence of viral DNA after 4 weeks and for disease symptoms after 6 weeks. Fig. 2(a) shows that, although the ability of the insects to transmit the virus decreased with age (from 100 to 50%), insects that acquired the virus after emergence showed a greater decrease in their ability to transmit TYLCV (from 100 to 10–20%). Therefore ageing alone could not account for the progressive decrease in virus transmission by adult insects that acquired TYLCV immediately after emergence.

The question of whether the ageing-related decrease in transmission was correlated with a decrease in virus acquisition was investigated. An experiment similar to that described above was performed. Following a 48 h acquisition access period on infected plants, insects were collected. Groups of insects were caged with tomato test plants for a 24 h inoculation access period and others were analysed for their TYLCV DNA content. Fig. 2(b) shows that after 10 days of age the amount of virus acquired by the insects started to decrease rapidly. At the age of 15 days, the insects acquired about half the amount of virus acquired by 10-day-old insects. At the age of 25 days, this amount was only about 10% and was barely detectable thereafter. The age-dependent decrease of virus acquisition was accompanied by a decrease in the ability to transmit the virus to test plants. It has to be noted that after the age of 25 days, at a time when viral DNA associated with the insects was barely detectable, the insects conserved about 50% of their ability to transmit TYLCV, although the appearance of viral DNA and symptoms in the inoculated test plants was considerably delayed compared to plants inoculated by younger insects.

**Effect of TYLCV on whitefly longevity**

The effect of lifetime association of TYLCV with *B. tabaci* on the longevity of the insects was assessed during four different periods in 1996–7: April–May, June–July, August–September and January–February. Adult whiteflies that emerged during a 24 h time period were caged with TYLCV-infected tomato plants for a 48 h virus acquisition access period. Control insects that emerged the same day were caged for 48 h with non-infected tomato plants. The two insect populations were then reared concurrently on egg-plants under exactly the same conditions in a nethouse. Climatic conditions were close to those found outdoors. The mortality rates of the two insect populations were compared.

The mortality curves displayed in Fig. 3, show that viruliferous as well as non-viruliferous individuals started to die at the age of approximately 10 days. In the 15–35-day time window, the mortality rate of infected insects was 1.5–1.8 times higher than that of non-infected insects, depending on the time the experiment was conducted (corroborated by *χ²* test, *P* < 0.0002). At the population level, the difference at the 50% mortality point between infected and non-infected insects was between 5 and 7 days: 27 vs 34 days in January–February, 20 vs 26 days in April–May, 26 vs 32 days in May–June and 29 vs 35 days in August–September. These results showed...
that the life expectancy of viruliferous insect populations was significantly lower than that of the non-viruliferous controls.

**Effect of TYLCV on whitefly fecundity**

Adult insects, 1 and 9 days after emergence, were caged with infected tomato plants for a 2 day acquisition access period (control insects of the same age were caged with non-infected plants). The 3- and 11-day-old viruliferous insects were then collected and caged with either egg-plants or tomatoes, using leaf cages (one insect per plant). The number of eggs laid by these insects during 1, 7 or 20 days was compared with those laid by the non-viruliferous insects.

The number of eggs laid by viruliferous and non-viruliferous insects as a function of oviposition time on egg-plants is shown in Table 1. The mean number of eggs laid during 1, 7 and 20 days on egg-plants by 3- and 11-day-old viruliferous and non-viruliferous insects is also shown in Table 1. The mean number of eggs laid by insects during the 24 h period following virus acquisition was similar to that laid by non-viruliferous insects, whether the insects were 3 days old (6.0 ± 0.94 vs 5.7, \( P = 0.4605 \)) or 11 days old (9.8 ± 0.79 vs 10.0, \( P = 0.8772 \)). The mean number of eggs laid by viruliferous insects during a 7 day period was significantly lower than that laid by non-viruliferous insects. This difference was more significant when the insects had access to infected plants when they were 9 days old (14.1 ± 28.0, \( P < 0.0001 \)) than when they were 1 day old (22.7 ± 38.1, \( P = 0.0583 \)). The mean number of eggs laid by 3-day-old viruliferous insects during a 20 day period

<table>
<thead>
<tr>
<th>Oviposition</th>
<th>Age of insect</th>
<th>Non-viruliferous insects</th>
<th>Viruliferous insects</th>
<th>Probability (( P ))</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 day</td>
<td>3 days</td>
<td>5.1 (0.75, ( n = 11 ))</td>
<td>6.0 (0.94, ( n = 7 ))</td>
<td>0.4605</td>
</tr>
<tr>
<td>11 days</td>
<td>10.0 (0.79, ( n = 10 ))</td>
<td>9.8 (0.79, ( n = 12 ))</td>
<td>0.8772</td>
<td></td>
</tr>
<tr>
<td>7 days</td>
<td>3 days</td>
<td>38.1 (4.49, ( n = 11 ))</td>
<td>22.7 (4.29, ( n = 12 ))</td>
<td>0.0583</td>
</tr>
<tr>
<td>11 days</td>
<td>28.0 (2.33, ( n = 11 ))</td>
<td>14.1 (2.53, ( n = 12 ))</td>
<td>&lt; 0.0001</td>
<td></td>
</tr>
<tr>
<td>20 days</td>
<td>3 days</td>
<td>56.0 (2.55, ( n = 20 ))</td>
<td>33.4 (2.55, ( n = 20 ))</td>
<td>&lt; 0.0001</td>
</tr>
</tbody>
</table>

**Fig. 3.** Mortality of viruliferous and non-viruliferous insects. One-day-old insects were caged with infected tomato plants. Control insects were caged with non-infected plants. The insects were collected after 48 h and caged with egg-plants using leaf cages (10 insects per plant, 15 plants for each insect population). The number of viruliferous (■) and non-viruliferous (●) insects (living versus dead) was counted every 3–4 days.

**Table 1. Effect of oviposition time on the number of eggs laid by viruliferous and non-viruliferous insects on egg-plants**

One- and 9-day-old insects were allowed to acquire virus during a 48 h access period on infected plants; non-viruliferous insects of the same age had access to non-infected tomato plants. The table shows the mean number of eggs laid during 1, 7 or 20 days by 3- and 11-day-old insects (the number of eggs laid by 11-day-old insects during a 20 day period is not included because of the high mortality of the ageing insects). The standard errors and the number of insects are shown in parentheses.

<table>
<thead>
<tr>
<th>Oviposition</th>
<th>Age of insect</th>
<th>Non-viruliferous insects</th>
<th>Viruliferous insects</th>
<th>Probability (( P ))</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 day</td>
<td>3 days</td>
<td>5.1 (0.75, ( n = 11 ))</td>
<td>6.0 (0.94, ( n = 7 ))</td>
<td>0.4605</td>
</tr>
<tr>
<td>11 days</td>
<td>10.0 (0.79, ( n = 10 ))</td>
<td>9.8 (0.79, ( n = 12 ))</td>
<td>0.8772</td>
<td></td>
</tr>
<tr>
<td>7 days</td>
<td>3 days</td>
<td>38.1 (4.49, ( n = 11 ))</td>
<td>22.7 (4.29, ( n = 12 ))</td>
<td>0.0583</td>
</tr>
<tr>
<td>11 days</td>
<td>28.0 (2.33, ( n = 11 ))</td>
<td>14.1 (2.53, ( n = 12 ))</td>
<td>&lt; 0.0001</td>
<td></td>
</tr>
<tr>
<td>20 days</td>
<td>3 days</td>
<td>56.0 (2.55, ( n = 20 ))</td>
<td>33.4 (2.55, ( n = 20 ))</td>
<td>&lt; 0.0001</td>
</tr>
</tbody>
</table>
was significantly lower than that laid by non-viruliferous insects (56±0 vs 33±4, P < 0.0001).

Table 2 shows the effect of the virus on the number of eggs laid on egg-plant and tomato during 7 days by insects that acquired virus when they were 1 and 9 days old. The mean number of eggs laid by non-viruliferous insects on egg-plant and on tomato was not significantly different (38±1 vs 36±5, P = 0.8242 for 3-day-old insects; 28±0 vs 32±5, P = 0.3572 for 11-day-old insects). The same was true for viruliferous insects (22±7 vs 28±0, P = 0.3441 for 3-day-old insects; 14±1 vs 12±9, P = 0.5808 for 11-day-old insects). Therefore, the host plant did not have a significant effect on the insect fecundity. Age had a significant effect on fecundity of viruliferous insects, but not on that of non-viruliferous insects. Three-day-old and 11-day-old non-viruliferous insects laid a similar number of eggs on tomato (36±5 vs 32±5, P = 0.4884) or on egg-plant (38±1 vs 28±0, P = 0.1485). On the other hand, the mean number of eggs laid by 11-day-old viruliferous insects was significantly lower than that of the 3-day-old viruliferous insects, on both tomato (12±9 vs 28±0, P = 0.0007) and egg-plant (14±1 vs 22±7, P = 0.0346). The total number of eggs laid on both tomato and egg-plants by viruliferous insects was significantly lower than that laid by non-viruliferous insects, whether the insects were 3 days old (24±8 vs 37±2, P = 0.0075) or 11 days old (13±5 vs 29±9, P < 0.0001).

The effect of TYLCV on the development of the whitefly egg was investigated. Whiteflies which had emerged during a 24 h period were caged with infected tomato plants for 2 days. The insects were then transferred to egg-plants (one insect per plant). The numbers of non-hatched eggs, empty eggs (ghosts) and crawlers (first and second instar) were counted after 20 days. The mean number of eggs laid by viruliferous insects during the 20 day period was significantly lower than that laid by the non-viruliferous control insects (33±4 vs 56±0, P < 0.0001). The percentage of eggs that developed into instars was similar, whether they were laid by infected or non-infected insects (9±9% vs 10±2%). Therefore, the virus influenced the number of eggs laid but not the emergence of the instars.

### Discussion

The relationship between TYLCV and *B. tabaci* appears to be more complex than thought before. We have investigated the long-term association of TYLCV from Israel with *B. tabaci* from a local colony. It has been mentioned that one of the factors affecting development, survival and fecundity of whiteflies is the host plant (Coudriet *et al.*, 1985; Gerling *et al.*, 1986). Therefore our measurements were made on insects reared on egg-plants of the same cultivar and of the same age, in a nethouse, under conditions close to those outside. The results presented here show that the passage of TYLCV in its vector is not neutral. TYLCV has several features reminiscent of an insect pathogen: (1) TYLCV DNA is retained in the insect during its entire adult life, (2) the virus significantly shortens the life-span of the insect, and (3) the virus has a negative effect on the insect fecundity.

The association of TYLCV with *B. tabaci* during the entire adult life of the insect was studied. One-day-old insects acquired virus during a single 48 h acquisition period on infected tomato plants. To avoid possible effects of disease-related plant metabolites on the insect, the tomato plants were infected 3 weeks before caging with insects, and presented barely detectable symptoms. The viruliferous insects were then reared on egg-plants (a TYLCV non-host), as were control whiteflies of the same age after the latter fed for 48 h on non-infected tomato plants.

TYLCV was associated with the insect during its entire adult life. While the viral DNA was detected during the insect’s life-span, the capsid protein was no longer detectable after 12 days. In parallel, the capacity of the viruliferous insects to transmit the virus regularly decreased, but did not disappear entirely. These results suggest that a considerable proportion of the virus ingested during the first 48 h after emergence progressively leaves the acquisition/transmission path and is stored in the insect cells, possibly in the form of nucleoproteins (Abouzid *et al.*, 1988). The decrease with time in the capacity of the viruliferous insects to transmit the virus was not solely

### Table 2. Effect of TYLCV on insect fecundity

<table>
<thead>
<tr>
<th>Age of insect</th>
<th>Host plant</th>
<th>Non-viruliferous insects</th>
<th>Viruliferous insects</th>
<th>Probability (P)</th>
</tr>
</thead>
<tbody>
<tr>
<td>3 days</td>
<td>Egg-plant</td>
<td>38±1 (4±49, n = 11)</td>
<td>22±7 (4±29, n = 12)</td>
<td>0.0538</td>
</tr>
<tr>
<td></td>
<td>Tomato</td>
<td>36±5 (4±13, n = 13)</td>
<td>28±0 (5±26, n = 8)</td>
<td>0.0538</td>
</tr>
<tr>
<td></td>
<td>Tomato + egg-plant</td>
<td>37±2 (2±99, n = 24)</td>
<td>24±8 (3±27, n = 20)</td>
<td>0.0075</td>
</tr>
<tr>
<td>11 days</td>
<td>Egg-plant</td>
<td>28±0 (2±33, n = 11)</td>
<td>14±1 (2±23, n = 12)</td>
<td>&lt; 0.0001</td>
</tr>
<tr>
<td></td>
<td>Tomato</td>
<td>32±5 (2±74, n = 8)</td>
<td>12±9 (2±15, n = 13)</td>
<td>&lt; 0.0001</td>
</tr>
<tr>
<td></td>
<td>Tomato + egg-plant</td>
<td>29±9 (1±77, n = 19)</td>
<td>13±5 (1±54, n = 25)</td>
<td>&lt; 0.0001</td>
</tr>
</tbody>
</table>
due to ageing of the insect, but was correlated with the long-term presence of the virus. Viruliferous insects of more than 3 weeks of age retained only 10–20% of their capacity to transmit the virus, compared with about 50% for non-viruliferous insects of the same age. The progressive loss of virus transmissibility by non-viruliferous insects was correlated with a decrease in virus acquisition.

The presence of TYLCV in insects was correlated with a significant decrease in life expectancy of viruliferous insects. The half-life of the viruliferous insect population was 5–7 days shorter than that of the non-viruliferous counterpart (a decrease of 17–23% in life expectancy), independent of the season in which the experiment was conducted. The half-life of the non-viruliferous population varied, from 26 days (in April–May) to 35 days in (August–September). Our values may be compared to those of Gerling et al. (1986) who found that longevity of B. tabaci adults in the field was 10–15 days during summer (temperatures in the high twenties), and 30–60 days in winter (temperature around 10 °C). To our knowledge, the effect of a plant virus on the longevity of its insect vector has never been thoroughly measured. Cohen et al. (1987) mentioned that healthy whiteflies better survived exposure to high temperature (30 °C for 4 h) than viruliferous insects (66-3% vs 46-4%, at 50% relative humidity; 78-3% vs 69-7% at 100% relative humidity).

As for other parameters of B. tabaci bionomics, fecundity values differ greatly with the experimental conditions. Gerling et al. (1986) indicated that oviposition rates on cotton average approximately 10 eggs/day at temperatures between 25 and 30 °C and decrease below 20 °C. Gameel (1974) claimed that maximum oviposition of B. tabaci occurs within the first week of adult life. We are not aware of any investigation of the effect of a geminivirus on the fecundity of its whitefly vector. We have studied the effect of TYLCV on insect fecundity. The plant on which eggs were laid, whether tomato or egg-plant, did not influence the number of eggs laid by the insects, viruliferous or not. We found that the association of TYLCV was correlated with a significant decrease in the fecundity of infected insects, measured as the mean number of eggs laid during a given period of time after virus acquisition. The negative effect of the virus was not immediately evident. The number of eggs laid during the first day after 48 h of virus acquisition by 1- or by 9-day-old insects was similar to that by non-infected insects of the same age. The negative effect of the virus on insect fecundity was evident when the oviposition periods were 7 or 20 days. This increasing negative effect with time may be related to the progressive invasion of many tissues by the virus, in particular invasion of the reproductive system. Indeed we found TYLCV DNA in eggs of infected insects (unpublished observations). The mean number of eggs laid per day by non-viruliferous insects decreased with age. It was 5-1 from 3 to 4 days of age, 5-4 from 3 to 10 days, and 2-8 from 3 to 23 days. This age-dependent decrease in fecundity was much more pronounced when insects acquired the virus for 2 days immediately after emergence. It was 6-0 from 3 to 4 days of age, 3-2 from 3 to 10 days, and 1-7 from 3 to 21 days.

Once the eggs were laid, the virus had no significant effect on the development of the immatures, although mortality rates were very high: only approximately 10% of the eggs, whether laid by infected or non-infected insects, developed into instars (measured in winter and in spring). It has been mentioned by Horowitz et al. (1984) that, during normal whitefly development, most mortality occurs in the egg and first larval stages. In a comprehensive life table for B. tabaci on cotton in the field, these authors concluded that mortality of the insect is highest during the crawler and young nymphal stages: only an average of about 17% and a maximum of 40% of the eggs reached the adult stage. The mortality of eggs and crawlers contributed most of the total mortality.

The findings presented in this paper indicate that TYLCV has many features of an insect pathogen. Virus–insect interactions should be taken into account in an integrated pest management programme. It would be possible to disturb the insect–virus interaction by enhancing, for example, the deleterious effects of the virus.

We thank Dr Bruno Gronenborn for the antiserum against the TYLCV capsid protein. This research was supported in part by grant 95-168 from the United States–Israel Binational Science Foundation (B. S. F.).

References


Long-term association of TYLCV with its vector


Received 4 April 1997; Accepted 2 June 1997