

Short communication

***Solanum nigrum*: an indigenous weed reservoir for a tomato yellow leaf curl geminivirus in southern Spain**

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Abstract

A tomato yellow leaf curl geminivirus (TYLCV-AL), was first identified in tomato plants in Almeria, southern Spain in 1992. This virus is transmitted by the tobacco whitefly, *Bemisia tabaci* (Gennadius), and is presently infecting tomato crops throughout the south eastern region of Spain. *Solanum nigrum*, collected from a field in south east Spain and exhibiting leaf curl symptoms, was squash blotted onto nylon membrane and gave a positive signal when hybridised to a TYLCV-Is DNA probe. Laboratory tests showed *B. tabaci* to transmit TYLCV-AL from infected tomato plants to *S. nigrum* seedlings. The virus could then be acquired by *B. tabaci* and transmitted back from infected *S. nigrum* plants to tomato, inducing typical TYLCV symptoms. These results indicate the importance of *S. nigrum* as a weed host/reservoir for a TYLCV and its possible role in the spread of this virus within Europe.

Results and discussion

Tomato yellow leaf curl virus (TYLCV), has been recorded in many different countries (Czosnek et al., 1988; Markham et al., 1994), and is one of many geminiviruses acquired and transmitted by the tobacco whitefly, *Bemisia tabaci* (Gennadius) (Bedford et al., 1994a). Symptoms include stunting and distortion of plants, leaf curling, inter-vein yellowing and necrosis of older leaves. Many different whitefly-transmitted geminiviruses, infecting different plant species from around the world, readily infect tomato (*Lycopersicon esculentum*) plants, producing the typical TYLCV symptoms (Bedford et al., unpublished data). Many of the present TYLCV epidemics may have originated from these indigenous plant species and therefore, may be different geminiviruses. It is also possible that 'new' TYLCV epidemics, in some areas, are linked to the appearance of a polyphagous strain of *B. tabaci*, the B biotype (Bedford et al., 1993), which has enabled endemic viruses to spread from their original host plants to tomato crops. Within Europe, TYLCV has

been reported from Sicily (Crespi et al., 1995), Sardinia (Caciagli et al., 1995), Spain (Moriones et al., 1993) and more recently Portugal (Louro et al., 1996). The Sardinian isolate shows a high similarity to the Spanish isolate (Noris et al., 1994), yet the Portuguese isolate is different and more homologous to the Israeli isolate (Louro et al., 1996).

In Spain, TYLCV has spread within the covered tomato crops around the Almeria region and is presently a widespread problem where tomatoes are grown as a multi-seasonal crop. New tomato crops are almost certainly infected by viruliferous whiteflies moving within and between greenhouses. However, in Mazarron and Aguilas, within the region of Murcia, many field grown tomato plants also become infected with TYLCV. One such field was observed in Mazzaron in September 1996, where the grower had to replace the first three rows of tomato plants on one side of the field, due to their severity of TYLCV infection. This severity implied that the plants were infected at a very young stage, soon after planting and by viruliferous whiteflies ingressing the crop from one direction. The only like-

ly source of whiteflies and inoculum was from a crop of melon, harvested soon after the tomatoes had been planted. Although cucurbit crops have not been shown as susceptible to TYLCV, they are often a good host for *B. tabaci* (Bedford et al., 1994b). Within the field where the melon crop had been harvested, many stunted nightshade, *Solanum nigrum*, plants were found with severe leaf distortions. These plants also hosted colonies of *B. tabaci*.

A sample of symptomatic *S. nigrum* was taken for laboratory analysis. Plant tissue was squash-dot blotted onto a nylon membrane and hybridised to a radio-labelled probe (Maule et al., 1983; Rigby et al., 1977), produced from a full length clone of the Israeli isolate of tomato yellow leaf curl virus, TYLCV-Is. The membrane was washed in a solution of 2xSSC and 0.1% SDS at 65 °C, then exposed to autoradiograph film for 24 h. This produced a positive result, confirming that the *S. nigrum* sample was infected with a geminivirus similar to TYLCV.

Approximately 200 *B. tabaci* were caged onto a tomato plant (var. Kondine Red), that had been infected with TYLCV-AL by grafting infected scions of tomato, collected from southern Spain in 1995. After a 48 h acquisition access period (AAP), the whitefly were transferred to 6 seedlings of *S. nigrum* for a 48 h transmission access period (TAP). Whiteflies were then removed and plants fumigated with Propoxur (Octavius Hunt Ltd.). After 17 days, all the *S. nigrum* plants exhibited stunting and leaf distortion symptoms, similar to those seen on the field collected plants. A dot blot hybridisation test confirmed their infection.

Approximately 200 *B. tabaci* were then caged onto these infected *S. nigrum* plants for a 48 h AAP then transferred to 10 healthy tomato seedlings var. Kondine Red, for a 48 h TAP. After 15 days all tomato plants exhibited typical symptoms of TYLCV. Their infection was also confirmed by a dot blot hybridisation test.

This study has shown that *S. nigrum* plants presently growing within field crops of south eastern Spain, are hosting a tomato infecting, whitefly-transmitted geminivirus. It has also shown that *S. nigrum* is a field weed host and reservoir for the present TYLCV-AL epidemic in southern Spain. Sequence data will however, be needed to determine whether or not the whitefly-transmitted virus in the field collected *S. nigrum* is the same virus infecting tomato plants and if the field tomato virus is the same as that within the covered tomato crops of Almeria.

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References

- Bedford ID, Briddon RW, Brown JK, Rosell RC and Markham PG (1994a) Geminivirus transmission and biological characterisation of *Bemisia tabaci* (Gennadius) biotypes from different geographic regions. *Annals of Applied Biology* 125: 311–325
- Bedford ID, Briddon RW, Markham PG, Rosell RC and Brown JK (1993) A new species of *Bemisia* or biotype of *Bemisia tabaci* as a future pest of European agriculture. In: *Plant Health and the European Single Market*, D. Ebbles (ed), BCPC Monograph No. 54: 381–386
- Bedford ID, Pinner M, Liu S and Markham PG (1994b) *Bemisia tabaci* – Potential infestation, phytotoxicity and virus transmission within European Agriculture. Brighton Crop Protection Conference – Pests and Diseases 1994, 3: 911–916
- Caciagli P, Bosco D and Al-Bitar L (1995) Relationships of the Sardinian isolate of tomato yellow leaf curl geminivirus with its whitefly vector *Bemisia tabaci* Gen. *European Journal of Plant Pathology* 101: 163–170
- Crespi S, Noris E, Vaira A and Accotto GP (1995) Molecular characterisation of cloned DNA from a tomato yellow leaf curl isolate from Sicily. *Phytopathologia Mediterranea* 34: 93–99
- Czosnek H, Ber R, Antignus Y, Cohen S, Navot N and Zamir D (1988) Isolation of tomato yellow leaf curl virus, a geminivirus. *Phytopathology* 78: 508–512
- Louro D, Noris E, Verrati F and Accotto GP (1996) First report of Tomato Yellow Leaf Curl Virus in Portugal. *Plant Disease* 80: 1079
- Markham PG, Bedford ID, Liu S and Pinner MS (1994) The transmission of geminiviruses by *Bemisia tabaci*. *Pesticide Science* 42: 123–128
- Maule AJ, Hull R, Donson J (1983) The application of spot hybridisation to the detection of DNA and RNA viruses in plant tissues. *Journal of Virological Methods* 6: 215–224
- Moriones E, Arno J, Accotto GP, Noris E and Cavallarin L (1993) First report of Tomato Yellow Leaf Curl Virus in Spain. *Plant Disease* 9: 953
- Noris E, Hidalgo E, Accotto GP and Moriones E (1994) High similarity among the TYLCV isolates from the west Mediterranean basin: The nucleotide sequence of an infectious clone from Spain. *Archives of Virology* 135: 165–170
- Rigby PWJ, Dieckmann M, Rhodes C, Berg P (1977) Labelling deoxyribonucleic acid to a high specific activity *in vitro* by nick translation with DNA polymerase. *Journal of Molecular Biology* 113: 237–251