# Breeding network to create tomato varieties resistant to tomato yellow leaf curl virus (TYLCV)

H LATERROT INRA Station d'amélioration des plantes maraîchères BP 94 84143 Montfavet cedex France

### Breeding network to create tomato varieties resistant to tomato yellow leaf curl virus (TYLCV).

#### ABSTRACT

TYLCV disease causes serious damage to tomato crops. Chemical control alone, or associated with special cropping practices, is insufficient and must be complemented by genetic control. Two parent TYLCV-resistance source plants were discovered as early as 1970, Lycopersicon pimpinellifolium and L peruvianum. New programmes now aim at tapping the resistance of L chilense. A large-scale international breeding network provides tomato breeders with basic TYLCV-resistant plant populations.

### Sélection en réseau pour la création de tomates résistantes au virus du *tomato yellow leaf curl* (TYLCV).

#### RÉSUMÉ

La maladie due au TYLCV provoque d'importants dégâts dans les cultures de tomate. La lutte chimique seule, ou associée à des pratiques culturales spéciales, ne suffit pas et doit être renforcée par une lutte génétique. Deux géniteurs, sources de résistance, ont été retenus dès 1970, Lycopersicon pimpinellifolium et *L peruvianum*. De nouveaux programmes visent maintenant à exploiter la résistance de L chilense. Un réseau international de sélection très large offre aux sélectionneurs des populations de base porteuses de la résistance au TYLCV.

### Selección en red para la creación de tomates resistentes al virus del *tomato yellow leaf curl* (TYLCV).

#### RESUMEN

La enfermedad debida al TYLCV provoca importantes daños en los cultivos de tomate. La lucha química sola, o asociada a prácticas culturales especiales, no es suficiente y debe ser resforzada por una lucha genética. Dos genitores, fuentes de resistencia, fueron seleccionados desde 1970, Lycopersicon pimpinellifolium y L peruvianum. Unos nuevos programas tienden hoy día a explotar la resistencia de L chilense. Una red internacional de selección muy grande ofrece a los seleccionadores unas poblaciones de base portadoras de la resistencia al TYLCV.

*Fruits,* 1995, p 439-444 *Version française* : p 478-480 © Elsevier, Paris

#### KEYWORDS

*Lycopersicon esculentum*, plant breeding, geminiviruses, symptoms, pest control methods, genetic control, pest resistance, selection, research network.

### MOTS CLÉS

*Lycopersicon esculentum*, amélioration des plantes, géminivirus, symptôme, méthode de lutte antiparasite, lutte génétique, résistance aux organismes nuisibles, sélection, réseau de recherche. PALABRAS CLAVES Lycopersicon esculentum, fitomejoramiento, geminivirus, sintomas, métodos de control de plagas, control genético, resistancia a las plagas, selección, redes de investigación.

# the disease and its distribution

The tomato yellow leaf curl virus (TYLCV), a geminivirus transmitted by the *Bemisia tabaci* whitefly, causes spoon-like leaf curl, often associated with leaf yellowing. TYLCV inhibits the growth of plants which, depending on their development phase when the virus attacks, produce few small-sized fruit or none at all.

This disease, well established in the Middle East (MAKKOUK and LATERROT, 1983) was extensively studied in Israel as early as the 1960s, and still represents a limiting factor for crops in this region. It is also present in the eastern part of the Mediterranean Basin, in Sudano-Sahelian Africa and as far as Cape Verde and Southeast Asia. Over the last few years, it has reached Sardinia, Sicily, Calabria and, in 1992, the Mediterranean coast of Spain.

Use of the molecular DNA-TYLCV probe for the Israeli strain, obtained at the Rehovot Agricultural University, revealed that the same virus is established in the entire zone affected by the disease (CZOSNEK *et al*, 1990), but recent studies demonstrated that the viral genome is highly variable in this geographical area (CZOSNEK, 1992).

Studies using the same probe showed that all leaf curl symptoms found in the area affected by the disease were not attributable to TYLCV. In Central America and the Caribbean region, geminiviruses transmitted by *Bemisia tabaci* (BROWN and BIRD, 1992) also infest tomato crops. The French West Indies are now affected by these geminiviruses (HOSTACHY and ALLEX, 1993), but the symptoms produced can differ considerably from those caused by TYLCV.

# vector control

Insecticides, although necessary, are insufficient for controlling the *Bemisia tabaci* whitefly. It is crucial to alternate active ingredients that are applied in order to reduce the risk of selection for resistant strains.

The association of chemical control with other methods can be relatively efficient in various areas and different cropping contexts. On the Mediterranean coast of Turkey, contamination rates were considerably reduced by delaying the fall crop planting by 2–4 weeks, allowing farmers to plant tomatoes when the *Bemisia tabaci* populations were in recession.

Physical protection of nurseries and young plants with fine-mesh nets or nonwoven netting is on the increase, with good results in Israel, Lebanon, Cyprus and Italy. In Sicily, the quick reaction of farmers from the beginning of outbreaks and the combined efforts of some nursery plant producers and farmers growing tomatoes under plastic structures considerably reduced the impact of TYLCV as early as the second year after the virus began affecting the island.

# genetic control

Studies conducted in India, the Persian Gulf and Africa revealed differences in susceptibility between tomato cultivars. Large-scale experiments are under way on the best ones, particularly in Africa. They are determinate growth, compacthabit, fixed varieties with a short growing period. These cultivars seem to be able to adapt well to climatic changes. In addition, their resistance to various stresses indicates that they should react well to viral contamination.

Some of these cultivars, able to produce tomatoes even in the presence of TYLCV, have been bred in the United States for resistance to curly top virus, transmitted by a leafhopper (MARTIN and THOMAS, 1986).

Research for higher resistance levels started as early as the 1970s, especially in the Middle East. Wild *Lycopersicon* species, closely related to tomatoes, are the source of various types of resistance in presently cropped tomato varieties (LATER-ROT, 1989). TYLCV resistance has thus been sought among these species.

Various resistance levels were highlighted, and two resistance sources were selected with the assistance of the American University in Beirut (Lebanon):

- "LA 121" which belongs to *Lycopersicon pimpi-nellifolium*;

 - "CMV sélection INRA", a line derived from a cross between two different origins of *L peruvianum*.

From the first of these parent plants, the improved "Latyle" population, carrying partial resistance,

has been distributed (LATERROT and MAKKOUK, 1983). The second parent plant, not as closely related to cultivated tomato, was crossed with the latter and a large F2 seed population was obtained and widely distributed to breeders.

In Israel, breeding using a different L peruvianum origin produced a series of F1 hybrids of determinate growth, including "F1 TY 20" (PILOWSKY, 1990). They show partial resistance with conditional expression, and no expression at all under difficult and highly contaminated environmental conditions.

Other F1 hybrids were extensively tested in the Middle East and in other areas affected by TYLCV. Those most frequently mentioned, and which have existed long enough to be able to assess their performance, are:

- F1 Jackal = E438 (Sluis en Groot), F1 Top-21 (Clause), F1 Tyking and F1 Tyger (Royal Sluis) are determinate growth hybrids suitable for field cropping, without stakes;

- F1 Fiona = E437 (Sluis en Groot) is a high determinate growth hybrid;

- F1 Tydal and F1 Tygold (Royal Sluis) are indeterminate growth hybrids, interesting for staked outdoor or protected cropping.

These hybrids also have the advantage of being resistant to several other diseases which were considered serious before the appearance of TYLCV, or before the worsening of TYLCV epidemics. Research for resistance to this geminivirus should not conceal the threat represented by these other parasites and the importance of the corresponding resistances that have already been isolated, especially those concerning the tobacco mosaic virus (TMV), Fusarium oxysporum f sp lycopersici and Meloidogyne spp nematodes. The origin of the resistance in these hybrids was not revealed by those who found them, because they kept it secret!

New programmes now aim at tapping the very high level of resistance in Lycopersicon chilense species, highlighted in Israel (ZAKAY et al, 1990) and confirmed in ORSTOM1 studies (FAR-GETTE, 1991). Hybridizations between L chilense and cultivated tomatoes were carried out, and backcrosses aimed at improving the genotypes obtained have been conducted by various breeders.

L chilense resistance was proven efficient against the geminivirus affecting Florida (SCOTT and SCHUSTER, 1991), and seems to have also been successfully used against geminiviruses of other New World areas. In addition, the resistance of several of the above-mentioned F1 hybrids seems to be expressed under American conditions.

# the breeding network and its operations

After distribution of the first improved populations at the end of the 1970s, the range of circulated material and the collaboration network were progressively widened, particularly after 1984 with the support of CEE-DG XII contracts to create improved TYLCV-resistant populations.

The breeding network involves a basic group of nine partners, financially supported by CEE-DG XII, who work in six Middle East countries and three Sudano-Sahelian countries (appendix 1). Some "free partners" joined the network. In 1993, 25 virologist or tomato breeders of public institutes or private breeding firms joined the basic group to establish breeding plots with populations created at the INRA Avignon research centre (France).

Populations derived from crosses between cultivated tomatoes and seven parent plants of different origins, belonging to four Lycopersicon species, represent the basis of the breeding programme (LATERROT, 1990). The partners set up these populations in areas and at times of very serious natural TYLCV outbreaks. Some participants favour plant contamination by setting the populations to be sorted under protective structures, in which they introduce young tomato plants infected by the virus and carrying Bemisia tabaci.

Phenotypes are chosen among resistant plants and a first batch from the obtained seeds is used for the breeding programme of the participant breeder, while a second batch is transmitted to INRA Avignon. Half of the breeders receiving seeds of the populations to be sorted out send seeds of the bred-resistant plants back to the 1) ORSTOM: Institut français INRA Avignon centre.

The seeds received at INRA are used either for coopération, France). general intercrosses or backcrosses with cultivated tomatoes, after which the obtained hybrids are

de recherche scientifique pour le développement en

intercrossed. The populations created are then sent to the partners to be bred again. The programme is based on several populations, thus backcrossing and intercrossing are carried out at INRA with one or several populations, while the breeding partners work on one or several other populations.

# situation concerning improved populations

"Composite populations" were created from two improved populations based on different original parents. In the last few years, three population types have thus been created:

The "Chépertylc 92" population includes plants of determinate growth; it was created from 65 lines bred by seven partners, in seven countries, from the Chétylc population derived from *L cheesmanii* "LA1401", and from 65 lines bred by 12 partners, in nine countries, from the Pertylceg population derived from *L peruvianum* "CMV sélection INRA".

The "Pimpertylc 93" population is segregating for the growth type; it was created from 39 lines bred by 19 partners, in 11 countries, in the Pimhirtylc population derived from *L pimpinellifolium* "Hirsute", and from 38 lines bred by 16 partners, in eight countries, from the Pertylc population derived from *L peruvianum* "CMV sélection INRA".

The Chiltylc populations were obtained in different ways. From *L chilense* LA1969, the delicate hybridization with cultivated tomatoes and the first backcross were successful. Consequently, two different breeding schemes are presently being used:

- accelerated breeding was carried out in collaboration with the Institut des sciences végétales (ISV-CNRS, Gif-sur-Yvette, France), by screening resistant plants through TYLCV agroinoculation (KHEYR POUR *et al*, 1991); seeds from the fourth backcross were harvested during the 1994 summer (Chiltylc 94 population), and are now being distributed network-wide;

 a multilocation breeding programme was conducted within the network; partners used plants in the progeny of the first backcross (Chiltylc 92 population); the second backcross was carried out during spring 1994 with the first lines received at INRA; the intercross of plants derived from the second backcross was done during spring 1995 (Multichiltylc 95 population). The goal of this second procedure was to produce a population from a wider breeding base (multilocation). Breeding schemes for these populations are presented in appendix 2.

To establish a breeding base which breeders from TYLCV- (or close geminiviruses) and *Pseudomonas solanacearum*-affected countries can tap, populations were created with combined resistance to these two types of pathogenic agents.

# references

- Brown J, Bird J (1992) Whitefly transmitted geminivirus and associated disorders in the Americas and the Caribbean basin. *Plant Dis* 76 (3), 220-225
- Czosnek H (1992) TYLCV isolates from various regions of the world are unusually heterogenous. *Tomato Yellow Leaf Curl Newsletter* (2), 2-3
- Czosnek H, Navot N, Laterrot H (1990) Geographical distribution of Tomato Yellow Leaf Curl Virus. A first survey using a specific DNA probe. *Phytopath Medit* 29 (1), 1-6
- Fargette D (1991) Quelques propriétés de la résistance variétale à l'enroulement de la tomate. *In: Resistance of the Tomato to TYLCV,* Proc seminar EEC Contract DG XII-TS-2A-0055F (CD) partners, 4–7 September 1991, Montfavet, Avignon, France, 47-49
- Hostachy B, Allex D (1993) Un nouveau défi pour les maraîchers des Antilles françaises. Un géminivirus de la tomate transmis par *Bemisia tabaci. Phytoma, La défense des végétaux* 456, 24-28
- Kheyr Pour A, Bendahmane M, Matzeit V, Accotto GP, Crespi S, Gronenborn B (1991) Tomato yellow leaf curl virus from Sardinia is a whitefly-transmitted monopartite geminivirus. *Nucleic Acids Res* 19 (24), 6723-6769
- Laterrot H (1989) La tomate. Intérêt et utilisation des espèces sauvages pour la création variétale. *PHM, Rev Hort* 295, 13-17
- Laterrot H (1990) An EEC programme to improve the resistance of the tomato to tomato yellow leaf curl virus. *In: 11<sup>th</sup> meeting of the Tomato Working Group of Eucarpia*, Torremolinos, Malaga, Spain, 6-8 March, 1990, 31-36
- Laterrot H, Makkouk KM (1983) Selection for partial resistance to tomato yellow leaf curl virus (TYLCV). *Tomato Genet Coop Rep* 33, 4-5
- Makkouk KM, Laterrot H (1983) Epidemiology and control of tomato yellow leaf curl virus. *In: Plant Virus Epidemiology.* Oxford, UK (Plumb RT, Tresh JM, eds), Blackwell Scientific Publications, 315-321

- Martin MW, Thomas PE (1986) Levels, dependability, and usefulness of resistance to Tomato Curly Top disease. *Plant Dis* 70 (2), 136-141
- Pilowsky M (1990) Tolerance to Tomato Yellow Leaf Curl Virus derived from *Lycopersicon peruvianum. Plant Dis* 74 (3), 248-250
- Scott JW, Schuster DJ (1991) Screening of accessions for resistance to the Florida tomato geminivirus. *Tomato Genet Coop Rep* 41, 48-50
- Zakay Y, Navot N, Zeidan M, Kedar N, Rabinovitch H, Czosnek H, Zamir D (1990) Screening *Lycopersicon* accessions for resistance to tomato yellow leaf curl virus: presence of viral DNA and symptom development. *Plant Dis* 75 (3), 279-281

#### Appendix 1

Official INRA-France partners of the network breeding programme (Situation: January 1994)

Cyprus	N Ioannou Ministry of Agriculture and Natural Resources Agricultural Research Institute, Nicosia
Egypt	SE Moustafa Ministry of Agriculture, Vegetable Research Station Nady El Saed Str Dokki 12311, Giza
Israel	H Czosnek Hebrew University of Jerusalem, Faculty of Agriculture PO Box 12, Rehovot 76100
Jordan	M Kasrawi University of Jordan, Faculty of Agriculture Amman
Lebanon	Y Abou-Jawdah American University of Beirut Faculty of Agriculture and Food Sciences Aub PO Box 11 - 0236 Beirut
Mali	D Dembele Ministère de l'Agriculture Station de Recherches Agronomiques de Baguineda BP 30, Bamako
Senegal	EV Coly Institut Sénégalais de Recherches Agronomiques (ISRA) Centre pour le Développement de l'Horticulture (CDH) BP 3120, Dakar
Sudan	GA Dafalla University of Gezira, Faculty of Agriculture Science PO Box 20, Wad Medani
Turkey	K Abak University of Cukurova, Faculty of Agriculture Adana 01330

Appendix 2 Chiltylc population breeding schemes.

Momor verte × LA 1969

Tropiva 3 × F1 BC1 BC1 ⊕ Chiltylc 92

BC1 Screening Agroinoculation TYLCV

F1 Tyking × 1 plant from BC1

BC2 Screening Agroinoculation TYLCV

F1 Tyking × 5 plants BC3 BC3 ⊕ Chiltylc 93

BC3 Screening Agroinoculation TYLCV

F1 Fiona × 6 plants BC4 BC4 ⊕ Chiltylc 94

Chiltylc 92

TYLCV multilocale selection 12 fields / 9 countries

F1 Tyking × 25 lines BC2

BC2-I (Intercross of 220 plants)

#### **Multichiltylc 95**

The number of plants shown corresponds to the number of plants whose progeny was actually used for the programme follow-up.

BC = backcross

 $\oplus$  = selfing

I = intercross