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D'Onghia A.M., Djelouah K.

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# Occurrence and spread of Citrus Tristeza in the Mediterranean area

K. Djelouah and A. M. D'Onghia<sup>1</sup>

Tristeza is the most destructive virus disease of citrus, which affects trees of sweet orange, mandarin, grapefruit and other citrus cultivars. The disease is caused by citrus tristeza virus, an aphid-borne closterovirus, with a flexuous thread-like particles of 2000 nm in length and 11 nm in diameter (Bar Joseph and Lee, 1989).

The disease has destroyed millions of trees throughout the world; it occurs in most citrus producing areas and represents a tremendous threat to the citrus industry of the Mediterranean basin, where areas are extensively grown with the intolerant scion-sour orange combination (Roistacher, 1991).

It has probably originated in China and as most of citrus graft transmitted diseases, the spread is connected with human activity, especially vegetative plant propagation, grafting and pruning, a long history of cultivation and worldwide exchange of germplasm (Lovisolò, 1993).

Epidemics occurred in Spain and Israel, while in other countries (Albania, Algeria, Cyprus, Egypt, Italy, Lebanon, Morocco, Palestine, Tunisia, and Turkey) only few isolated foci of the disease have been discovered (Davino *et al.*, 1983; Bar Joseph and Lee, 1989; Kyriakou *et al.*, 1992; Bové, 1995; D'Onghia *et al.*, 1998; Stamo and D'Onghia, 1998; Jarrar *et al.*, 2000) (Table 1).

Apparently, all early cases of tristeza disease found in the citrus areas of the Mediterranean basin can be traced back to the introduction of in-

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<sup>1</sup> Istituto Agronomico Mediterraneo di Bari (Italy)

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fected budwood from abroad; all countries which have introduced the Meyer lemon have introduced tristeza as well, these includes Algeria, Cyprus, Israel, Italy, Morocco and Tunisia, (Bové, 1966). Other varieties, imported from Australia, South Africa, Japan, and the United States of America, have introduced tristeza into Mediterranean countries (Bové, 1995).

Spain is the first Mediterranean country where extensive spread of tristeza has occurred, more exactly in the district of Valencia, where it started in 1957; since then it has caused the death of about 15 million trees. Furthermore, the proximity of Morocco to Spain makes the threat of this virus to the Moroccan citrus industry now more than ever, a matter for concern (Bové, 1995).

More recently tristeza has been found in Israel; the virus could not be eradicated and has produced severe damages in certain areas. This outbreak of the disease might pave the way, in the near future to the spread of tristeza in orchards of the Jordan Valley and Syria, where the disease was not reported.

Table 1. Occurrence of Tristeza disease in different Mediterranean countries

Country	Situation	References
Albania	+	Stamo and D'Onghia, 1998
Algeria	+	Frezal, 1957
Cyprus	+	Mendel, 1956; Kyriakou <i>et al.</i> , 1993
Egypt	+	Nour Eldin and Bishay, 1958
France	+	Bové <i>et al.</i> , 1988
Greece	+	Dimou <i>et al.</i> , unpublished data
Israel	+	Raccah <i>et al.</i> , 1976
Italy	+	Davino <i>et al.</i> , 1983; D'Onghia and Savino Unpublished data
Jordan	-	
Lebanon	+	D'Onghia <i>et al.</i> , 1998
Libya	+	Nourelidin and Fudlallah, 1976

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Malta	-	
Morocco	+	Nhami and Kissi, 1978
Palestine	+	Jarrar <i>et al.</i> , 2000
Portugal	+	Nolasca, unpublished data
Spain	+	Moreno <i>et al.</i> , 1988
Syria	-	
Tunisia	+	Rebour 1950 <i>In</i> Bové, 1966
Turkey	+	Norman, 1963

+ reported,

- not reported

In Palestine, CTV has recently been detected only in the West Bank, and not in the Eastern part (Jarrar *et al.*, 2000). This suggests that the virus may have entered Palestine from Israel, either through infected propagating material or viruliferous aphids, or both. Indeed, like other Mediterranean countries, Palestinian citrus industry is mostly based on sour orange rootstocks, and therefore CTV represents a serious threat.

The disease is vector transmitted in a semi persistent manner (Bar Joseph *et al.*, 1979) by several aphid species whose efficiency of transmission is variable. The main vector is *Toxoptera citricida* which is not present in the Mediterranean basin and may adapt to various climatic conditions, spreading rapidly from country to country. Recently it has entered Madeira island (Cambra, comm. Person); where *Aphis gossypii* (Whiteside *et al.*, 1988) and *Aphis citricola* (Rocha Pena *et al.*, 1995) prevail in the Mediterranean area, other aphid species have been reported as vectors but with a lower efficiency as *T. aurantii*, *A. craccivora* and *Myzus persicae* (Bové, 1995).

Another alarming discovery was that tristeza virus can mutate, thus causing changes which might induce rapid spread by alternative aphids. In California, the severe seedling yellows form of tristeza was found to spread rapidly when carried by *A. gossypii* (Roistacher *et al.*, 1980). In Israel it was shown that this same aphid could transmit a particular tristeza virus strain at a rate of approximately 40

%, versus less than 5%, in case of two other strains (Bar Joseph and Loebenstein, 1973). Favourable climatic conditions, allowing the build-up of high aphid populations, as occurred in Spain after frost damages, may lead to the rapid dissemination of tristeza (Bové, 1995).

As reported by Bové (1995), the world citrus growing area may currently be divided into three categories in relation to tristeza.

*Category I:* this includes areas in which practically all citrus trees are infected with the virus, or where the virus is spreading rapidly, the vector being *T. citricida*.

*Category II:* this includes areas not yet infected by the virus, but in which most trees are susceptible to the disease and *T. citricida* is not present. Mediterranean countries except Spain, and Israel fall in this category. Sour orange is extensively used in these areas and isolated declining trees have been found to be infected with the virus. However, no evidence of field spread has been observed except for Spain and Israel.

*Category III:* this includes citrus areas where the tristeza virus is known to be spreading but the vector is not *T. citricida* e.g. Spain and Israel.

Spain: The virus is spreading in the area of Valencia Vectors are *T. aurantii*, *A. gossypii*; *Myzus persicae* and possibly, others; Israel: Rapid spread has occurred, *A. gossypii* is considered an active vector.

CTV infection causes a wide range of symptoms in citrus, depending on the environmental conditions, host species and virus isolates. The most common economically important symptoms are decline or death of trees grafted on sour orange rootstock and/or stem pitting of the scion of the rootstock. CTV infects nearly all the citrus species, whereas, the only known non-rutaceous host is *passiflora*.

Generally, mandarins are tolerant and trifoliolate resistant to CTV infection (Bové, 1995); some species i.e. Satsuma, Kumquat etc., are healthy carri-

ers of the disease and they represent a dangerous inoculum source for the virus spread, (Whiteside *et al.*, 1988). In addition to the typical tristeza disease, CTV can also induce stem pitting, a disorder that occurs in susceptible varieties regardless of the rootstock and the seedling yellows, a reaction experimentally induced by tissue graft inoculation of certain strains of the virus into young seedlings of sour orange, grapefruit, lemon and some citron (Roistacher, 1991).

Biological indexing on Mexican lime is one of the most effective techniques for CTV mass diagnosis. On the other hand, the use of enzyme labelled antibodies in serological assays has provided a valuable diagnostic tool, extremely sensitive, stable, cost-effective and safe (Bar Joseph *et al.*, 1980; Garnsey *et al.*, 1993, Rocha Pena and Lee, 1991), mass testing when tristeza symptoms are not clear; indeed, it's possible to detect mild forms of the virus that can be missed by indexing (Roistacher, 1991)

A number of different methods have been developed for CTV detection such as, sodium dodecyl sulfate (SDS) immunodiffusion (Brlansky *et al.*, 1984), serologically specific electromicroscopy (SSEM), SSEM gold labelled assay, enzyme linked immunosorbent ELISA (Bar Joseph *et al.*, 1979; Cambra *et al.*, 1991; Rocha Pena and Lee, 1991, Rocha Pena *et al.*, 1991), radioimmunosorbent assay RISA (Rocha Pena and Lee, 1991), *in situ* immunofluorescence (ISIFR) (Brlansky *et al.*, 1988), Western blot assay (Guerri *et al.*, 1990) dot immunoblot assay (Rocha Pena *et al.*, 1991) and direct tissue blot immunoassay DTBIA (Garnsey *et al.*, 1993). Recently, bacterially expressed coat protein fragments have been used to produce CTV specific serological tool (Nikolaeva *et al.*, 1995; 1998), moreover, molecular hybridisation has also been used (Rosner *et al.*, 1986).

CTV distribution in the Mediterranean countries is not at all reassuring, in that, most of these countries grow their citrus orchards with intolerable scion-sour orange combination, the inoculum is widely distributed and one of the most efficient CTV vector *Aphis gossypii* is present. As a result,

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preventive measures aimed at CTV eradication should urgently be implemented; to this end, a more extensive survey of citrus groves and nurseries in most Mediterranean countries is highly recommended to identify and immediately destroy the infected trees.

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