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Virus and virus-like diseases of citrus in Turkey

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SUMMARY - More than 15 virus and virus-like diseases were detected in Turkish citrus groves among which psorosis complex, stubborn, cachexia, exocortis, infectious variegation, concave gum, impietratura, cristacortis, satsuma dwarf, gummy bark and woody gall. Tristeza is present in a mild form and apparently it is not vector-transmitted in the field. A new disease transmitted by *Parabemisia myricae* has been reported, which is very similar to citrus infectious variegation.

Key words: citrus, viruses, virus-like, diagnosis, Turkey

RESUME - Plus de 15 maladies à virus et de type viral ont été détectées dans les vergers d'agrumes turques: le complexe de la psorose, le stubborn, la cachexie, l'exocortis, la panachure infectieuse, la concavité gommeuse, l'impietratura, le cristacortis, le nanisme du satsuma, l'écorce gommeuse et les galles du bois. La tristeza est présente sous une forme faible et apparemment, elle n'est pas transmise par vecteurs. Une nouvelle maladie transmise par l'aleurode *Parabemisia myricae* a été signalée qui s'avère être très similaire à la panachure infectieuse.

Mots-clés: agrumes, virus, virus similaires, diagnostic, Turquie

Citrus industry

Citrus production in Turkey is increasing year by year due to high demands for local consumption and exportation. The total citrus production is about 1.500.000 tons/year. According to this production, Turkey ranks tenth in the world and fourth in the Mediterranean. About 120,000 ha are given over to citrus (Anonymous, 1992a, 1992b).

Generally, because of its vigorous growth and compatibility with common scion cultivars, sour orange is widely used as a rootstock in the Eastern Mediterranean area of Turkey. Trifoliate orange is used as rootstock in the Aegean and Black Sea regions. Carrizo and Troyer citranges may be used as rootstocks by the Turkish citrus growers in the near future. The major citrus varieties are: orange (Navel oranges, especially Washington Navel, Valencia, Shamouti, Sanguinello and Moroblood), grapefruit (Marsh Seedless, Red Blush, Star Ruby, Rio Red, Henderson and Rey Ruby), lemon

(Interdonato, Kutdiken, Lamas, Meyer and some local varieties), mandarin (Satsuma, Fremont, Clementine and some hybrids, i.e. Lee, Nova and Robinson) and Minneola tangelo .

Similarly to many other citrus producing countries, the Turkish citrus industry is threatened by many virus and virus-like diseases. At present, 15 virus and virus-like diseases, e.g., psorosis complex, infectious variegation (crinkly leaf), chlorotic dwarf, tristeza, satsuma dwarf, cristacortis, concave gum, impietratura, gummy bark, woody gall, cachexia-xyloporosis, exocortis, and stubborn, have been reported in citrus. Sieve tube necrosis, bud union crease, and rumple are also found but their causal agents are unknown. It must be kept in mind that if a thorough investigation was carried out via advanced techniques, many more viruses are likely to be identified

Graft-transmissible diseases

- Virus and virus-like diseases

The **psorosis complex** consists of psorosis A, psorosis B, and ringspot (Roistacher 1993). Only psorosis A is reported in Turkey. This disease is widespread and may be found in all citrus producing areas of Turkey causing early death, reduced vigour and lower yields of trees. Appearance of bark scaling symptoms in trunk takes 10-12 years. Various varieties of sweet orange including Washington Navel, Shamouti, Valencia, and some local varieties were found to be infected. Psorosis was also observed in Dancy, Clementine, Fremont and Satsuma mandarins and Marsh seedless grapefruits. Psorosis diseases are widespread in Navel and Satsuma plantations. The incidence of psorosis complex varies from 64 % to 80.5 % in Navel oranges and 15.8 % to 31.0 % in Satsuma mandarin in the Eastern Mediterranean region (Gullu, 1989) and 50 % in Satsuma mandarin in the Aegean region (Azeri and Heper, 1973). However, in the Eastern Black Sea region, the incidence was 1-2 % (Cengiz *et al.*, 1968).

Tristeza is a very destructive disease of mandarin, grapefruit and sweet orange grafted on sour orange and causes stunting, slow dieback or sudden decline, defoliation and death of the tree. Tristeza induces an overgrowth above the bud union and starch depletion in the rootstock portion of the tree (Salibe, 1986). The possible presence of tristeza in Turkey was first reported by Norman (1963), CTV was also reported in the Aegean Region (Ozalp and Azeri 1967), in Çukurova region (Cengiz *et al.* 1976). CTV was determined by biological indexing and serological assays in the Eastern Mediterranean region (Baloglu, 1988). Azeri and Heper (1973) estimated that the incidence of CTV is about 16% in Satsuma mandarins in the Aegean region. Cinar *et al.* (1993) reported that it is 0.5 % in Navel oranges and 0.04 % in Satsuma mandarins in the Çukurova region.

Satsuma dwarf virus causes dwarfing, multiple sprouting, little leaf and chlorotic symptoms on the leaves of Satsuma mandarins (Tanaka, 1968). This disease was first reported in Satsuma mandarin trees (Azeri and Heper, 1973). The virus incidence was found to be about 2 % in the Aegean region but in the Çukurova region the virus incidence was about 31.6% in Satsuma mandarins (Gullu, 1989).

Concave gum causes broad concavities, especially on the trunk and main branches of mandarin, grapefruit, sweet orange and lemon, sometimes with gumming through cracks in the bark and "oak leaf" pattern in the young leaves (Roistacher 1993). Concave gum is also widespread in the Eastern Mediterranean region of the country. Since concave gum used to be included in the psorosis complex, a separate survey has never been made for this disease. However the incidence of concave gum and psorosis A was reported equal to 47 % in the Navel group; whereas 18% of Satsuma mandarin trees were infected with concave gum and psorosis A (Cinar *et al.* 1993).

Impietratura disease affects grapefruit, orange and lemon fruits, and has been found in many citrus producing countries of the Mediterranean basin. The disease causes small and hard fruits with gumming and browning (Klotz, 1973, Norman, 1963). Impietratura was first reported by Chapot (1961) in Turkey. The incidence of the disease was found to equal 80.3 % in grapefruit and 28.9 % in local orange varieties in the Çukurova region (Nas, 1989). Recently, we have observed impietratura symptoms in newly-established *Minneola tangelo* orchards.

Cristacortis symptoms are similar to cachexia-xyloporosis causing stem pitting symptoms without gumming in the trunk or bark of scion and rootstock (Salibe, 1986). According to Roistacher (1991) the disease can be distinguished by sharp and deep pitting in the tangelo and mandarins with gum usually at the base of the pits. It was first reported by Moreira (1965) in Shamouti orange trees, in Antalya. Azeri (1979) reported the occurrence of cristacortis (20 %) in mandarin trees in Bodrum-Izmir. According to Gullu (1989) the incidence of cristacortis in the Çukurova region is about 0.4 % .

Gummy bark was found by Moreira (1965) in orange trees its incidence ranging from 1.2 % to 48.4% in Navel trees budded on sour orange in the Çukurova region (Gullu, 1989). Infected trees show stunting, wood pitting in the trunk and gum impregnation above the bud union (Salibe, 1986). Recently, it was reported that 28% of Navel oranges are infected with gummy bark disease of citrus in the Eastern Mediterranean region (Gullu, 1989).

• Viroid diseases

Cachexia disease, also known as xyloporosis, was reported by Reichert (1959), Norman (1963) and Moreira (1965) in Turkey. It induces gum impregnation of the phloem and cambial pitting of the wood with corresponding pegging of the bark on mandarins, tangors, tangelos, *Citrus macrophylla*, *Citrus volkameriana* and other varieties (Calavan and Christiansen, 1965). Cachexia-xyloporosis is a widespread disease in most old mandarin orchards. It was observed in *Minneola tangelo* and different grapefruit varieties. The incidence of cachexia- xyloporosis-affected Satsuma mandarin trees in the Eastern Mediterranean region is about 21.6 % - 64.4 % (Gullu, 1989), 1-6 % in the Eastern Black Sea region (Cengiz *et al.*, 1968) and 1.58 % in the Aegean region (Azeri and Heper, 1973). Recent studies showed that 50 % of Satsuma and Clémentine mandarins are infected with cachexia-xyloporosis (Cinar *et al.* 1993).

Exocortis causes bark splitting, scaling and gumming of the rootstock portion of the trunk and stunting. The disease affects trifoliolate orange and most of its hybrids, Rangpur lime, Palestine sweet lime (Fraser *et al.*, 1961; Salibe, 1986). Exocortis disease was reported by Norman (1963) and by Moreira (1965) on trifoliolate rootstock and hybrids in Turkey. Later, Cengiz *et al.* (1976) reported that Moro, Shamouti, and Washington Navel oranges, Interdonato, and Femminello lemons, Satsuma mandarins and other varieties grafted on trifoliolate rootstock were found to be infected with exocortis. Azeri and Heper (1973) reported that 4.26% of Satsuma mandarins were infected with exocortis and 1.58% with cachexia in the Aegean region. Cachexia (CVIIb), exocortis (CEV), CVIa, CVIb, CVIIa, CVIII or CV IV viroids were identified in Çukurova region by PAGE (Onelge, 1994). According to Cinar *et al.* (1993) 15 out of 16 navel orange trees and 10 out of 12 Satsuma mandarin trees were tested positive for exocortis. Commercial citrus orchards are probably entirely infected with exocortis. However, due to the use of sour orange as rootstock, symptoms are not visible in the field.

- Prokaryote-induced diseases

Stubborn disease of citrus, caused by *Spiroplasma citri*, is transmitted by leafhoppers and causes stunting, bushy appearance, numerous short leafy branches, growth of axillary buds, premature leaf drop, twig dieback, off-season flowering, excessive fruit drop; acorn-shaped or lopsided fruits with curved columella, small fruits and low yields (Fawcett and Klotz, 1948; Calavan, 1969; Salibe, 1986). Sweet oranges, particularly Washington Navel, Fremont mandarin, grapefruit and tangelo, are very susceptible to stubborn. Stubborn is a serious disease of citrus in the Mediterranean region and Near East (Bové, 1986 ; Caglayan, 1987; Cinar *et al.* 1993). Due to active vector transmission, 50 % of Navel orange trees in the Çukurova region are infected with stubborn (Gullu, 1989), which was also observed in Navel oranges and in Satsuma mandarin in the Aegean region (Azeri and Heper, 1973; Azeri, 1987). No other intracellular prokaryotes have been so far found in Turkish citrus.

- Other disorders

Several disorders were observed showing symptoms ascribable to virus diseases, e.g. sieve tube necrosis and rumple in lemon. However, the causal agents of these disorders are still unknown.

New diseases

Citrus chlorotic dwarf (CCD), previously believed to be a crinkle leaf type disease (Korkmaz *et al.*, 1994), is widespread in Mersin and causes crinkled and puckered leaves in lemon, tangelo and orange trees. It has recently become epidemic especially in Kutdiken lemon (a common local variety) and Minneola tangelo. Korkmaz *et al.* (1994) reported CCD as a new whitefly-borne virus-like disease which is transmitted from citrus to citrus by the whitefly *Parabemisia myricae*. The disease symptoms are similar to those induced by citrus infectious variegation except that the infected plant in the field shows smaller leaf size. Another crinkle-type infectious agent was mechanically transmitted to herbaceous plants (Baloglu *et al.* 1992; Sen and Baloglu, 1994). Martelli *et al.* (1994) isolated olive

latent virus-1 from CCD-infected Kutdiken lemon and Mexican lime. It could be possible that more than one virus is involved in the induction of crinkling type of symptoms.

Research on citrus diseases

The first attempt to detect citrus virus diseases in Turkey started in 1950. Stubborn and impietratura diseases were first reported in the Eastern Mediterranean region by Chapot (1959; 1961). Later, Norman (1963) reported the occurrence of exocortis, cachexia-xyloporosis and psorosis diseases. He also pointed out the importance of tristeza virus.

In the last three decades several virologists have worked with citrus virus and virus-like diseases in the country (Chapot, 1961; Norman, 1963; Azeri and Heper, 1973; Azeri, 1979; Caglayan 1987; Baloglu, 1988; Gullu, 1990; Cinar *et al.*, 1993; Onelge, 1994). Some vector-virus relations were also investigated especially for aphid vectors of tristeza, whitefly vectors of CCDV and leafhopper vectors of stubborn (Yilmaz *et al.* 1990; Kersting and Sengonca, 1992; Korkmaz, *et al.* 1994).

Several methods have been used for the identification of citrus viruses in Turkey. Symptomatological observations in the field (Table 1) are generally used as a diagnostic tool. However, some pathogens do not cause visible symptoms. Therefore, biological indexing, serology, PAGE, cultural assay and Electron microscopy are also used in diagnosis (Table 1). Biological indexing is the major diagnostic method. The indicator plants used are listed in Table 2. The research activities for the diagnosis of citrus virus and virus-like diseases are carried out at the Department of Plant Protection, and the Subtropical Fruits Research and Experimental Centre of the Çukurova University, Adana, at the Regional Plant Protection Research Institute in Adana, at the Greenhouse Farming and Citrus Research Institute in Antalya and at the Regional Plant Protection Institute of Bornova in Izmir.

Conclusion

More than 75 virus and virus-like diseases have been reported in citrus all over the world (Salibe 1986), but only 15 of them have so far been found in Turkey. For example, citrus variegated chlorosis, greening disease and some destructive strains of CTV (as reported from Brazil, California, Florida, Australia, Venezuela, Israel and Peru) have not been found as yet.

It is probable that if advanced techniques are used for diagnosis and a detailed survey conducted in all Turkish citrus areas, other agents can be detected. Because of the introduction of propagation material from abroad, graft-transmissible viruses of citrus were introduced in the country in the past. Therefore, more attention should be paid :

- To strengthen quarantine measures for the control of the introduced material.
- To have a legislation for the mandatory certification programmes of citrus.
- To establish an advanced laboratory for an early detection of diseases.

- To have periodical training courses on the "application of new detection techniques" in the citrus producing countries.
- To promote collaboration with the researchers of other citrus producing countries (at least between Mediterranean countries) for quarantine regulations.
- To establish rules for a Mediterranean certification programme.

Table I - Field symptoms and diagnostic methods of virus and virus-like diseases of citrus

<i>Disease</i>	<i>Symptoms in the field</i>	<i>Diagnostic methods</i>
Psorosis A	Bark scaling on sweet orange, mandarin, grapefruit and leaf symptoms in spring	Indexing
Blind pocket	Deep and narrow concavities in trunk of sweet orange, mandarin etc.	Indexing
Concave gum	Oak leaf pattern, concavities in the trunk, gum formation under bark of sweet orange, mandarin, tangerine and lemon	Indexing
Infectious variegation or Crinkly leaf	Leaf crinkle , distorted leaves with variegation in lemon, mandarin, sweet orange, grapefruit.	Indexing, Mechanical transmission and ELISA
Chlorotic Dwarf	Distorted leaves, puckering, flecking, dwarfed leaves in sensitive varieties	Indexing
Satsuma dwarf	Severe dwarfing of the trees	Indexing, Mechanical transmission and ELISA
Impietratura	Gum spots in fruit albedo and oak-leaf symptoms	Indexing
Tristeza	Gradual decline, dieback, inverse pitting below bud union on sweet orange, mandarin, grapefruit on sour orange rootstock	Indexing and ELISA
Cachexia-xyloporosis	Gum pockets and pits in bark of mandarin plants	Indexing and Electrophoresis
Exocortis	Stunting and scaling of the bark of trifoliolate rootstock, no symptoms expression in sour orange	Indexing and Electrophoresis
Cristacortis	Pits in scion and rootstock, oak leaf symptoms in spring flush of mandarin and sweet orange	Indexing
Gummy bark	Gum outbreaks on sweet orange	
Stubborn	Small and spoon shaped leaves, irregular blooming time, acorn shaped fruit, seed abortion	Indexing, ELISA and Culture assays

Table II - Indicator plants used for citrus virus and virus-like diseases

Disease	Indicator plants	Symptoms
Psorosis complex	Pineapple sweet orange , Dweet tangor	Shock reaction, oak leaf pattern, vein banding, vein clearing
Crinkly leaf or Infectious variegation	Etrog citron, Mexican lime, Sour orange	Crinkling, variegation and cupping of leaves
Concave gum	Dweet tangor	Oak leaf pattern, internerval flecking
Chlorotic Dwarf	Rough lemon	V-shaped inlet on very young leaves, diffuse flecking
Tristeza	Mexican lime, Sour orange Grapefruit	Vein clearing, and cupping of leaves.
Satsuma dwarf	White sesame	Necrotic local lesions on primary leaves and epinasty in secondary leaves
Cachexia-xyloporosis	Parson's special mandarin on rough lemon	Gum formation over the bud union in the bark of the scion (when the bark peel)
Exocortis	Etrog citron (861 S-1 selection)	Leaf epinasty and browning of mid-vein and petioles
Cristacortis	Orlando tangelo, Dweet tangor	Pits on tangelo plant, oak leaf pattern
Stubborn	Madam Vinous sweet orange	Stunting, small leaves and cupping of leaves, chlorotic spots in the leaves

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