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Virus and virus-like diseases of citrus in Italy with special reference to Apulia

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SUMMARY - A general view on the sanitary status of citrus in Apulia is given based on testing of 1750 plants from commercial groves and nurseries. Early varieties of clementine ('Common' and local ecotypes) and sweet orange ('Navelina') are the most important ones in the region but they are highly infected by virus and virus-like diseases. Cachexia and exocortis are the most widespread diseases followed by concave gum, psorosis complex and cristacortis. Citrus infectious variegation and impietratura are rarely detected. No evidence of quarantine pests has been reported with the exception of CTV, detected in an ornamental citrus and in a Chinese lemon.

Key words: citrus, viruses, virus-like, diagnosis, Apulia, Italy

RESUME *- Une vue d'ensemble de l'état sanitaire des agrumes dans la région des Pouilles est présentée en prenant en considération les résultats des tests exécutés sur 1750 plantes provenant des vergers commerciaux tout comme des pépinières. Les variétés précoces de clémentinier (des écotypes "Commun" et locaux) et d'orangers doux ("Navelina") sont les plus importantes dans la région, mais elles sont considérablement infectées par des maladies à virus et de type viral. Les viroïdes de la cachexie et de l'exocortis sont les plus répandus, suivis de la concavité gommeuse, du complexe de la psorose et de la cristacortis. La panachure infectieuse et l'impietratura ne sont détectées que rarement. Il n'existe pas non plus de signalements d'agents de quarantaine, à l'exception d'un citrus ornemental et d'un citronnier chinois qui se sont révélés infectés par le CTV.*

Mots-clés*: agrumes, virus, virus similaires, diagnostic, Pouilles, Italie*

Citrus industry in Apulia

The Italian citrus industry, apart from Sardinia, Lazio and Liguria, is mainly concentrated in the southern regions in virtue of the suitable soil and climatic conditions for the cultivation of the various species which prefer tropical and subtropical regions. The surface area and productions for each citrus species in the main citrus-growing regions are reported in Table I. Apulia ranks third after Sicily and Calabria, representing the northernmost limit of the citrus-growing area. In Apulia citrus trees are

cultivated along the Ionian coast and, to a lesser extent, in Gargano and Salento thus covering about 10,000 ha of cultivated surface (Table I).

Table I - Area and production of citrus in the main Italian regions (ISTAT, Annuario 1995)

Main citrus species	Area(ha)	Production (ql.) ql/ha	Total
Sicily			
Sweet orange	65,732	220.4	14,354,265
Mandarin	7,947	158.9	1,256,730
Clementine	4,875	181.4	867,369
Lemon	34,115	215.3	7,330,202
Calabria			
Sweet orange	26,661	259.9	6,762,078
Mandarin	2,340	196.4	445,730
Clementine	8,972	217.9	1,670,826
Lemon	1,342	202.4	266,930
Apulia			
Sweet orange	5,770	145.0	519,518
Mandarin	428	170.2	67,055
Clementine	3,500	133.1	401,145
Lemon	307	136.7	37,732
Italy			
Sweet orange	109,797	222.0	23,426,433
Mandarin	12,626	164.9	2,033,395
Clementine	20,573	180.4	3,297,332
Lemon	38,330	211.8	8,021,044

Most of citrus productions are of an excellent quality and they are represented by blond sweet oranges, especially navel group ('Navelina', 'Washington Navel'), mandarins ('Avana' and 'Tardivo di Ciaculli') and clementines, which has fully replaced the mandarin ('Common', 'Oroval', 'Nules' and local ecotypes 'Fedele', 'Spinoso', 'Precoce di Massafra' and other mutations) (Table II). The major interest is however paid to local early ecotypes of clementine, which ripen at the beginning of October, giving high profits to growers. Satsuma mandarin was recently introduced so as to cover an empty citrus market period in September, as well as tangelo cvs. Minneola and Nova. Tangelos were initially very successful, due to their utilization for juice production. Scarcely represented are lemon, grapefruit, and some other citrus trees since Apulia is a cold region for their cultivation.

Early varieties are the most important in Apulia to avoid the risks of late spring frost; nonetheless, the variety collection includes late ecotypes (mandarin and oranges) to allow a better distribution of productions which are in surplus in some short periods of the year.

Almost all citrus trees are grafted onto sour orange rootstock because of its resistance to *Phytophtora spp.* and its better adaptation to the soil conditions.

The regional nursery situation, similarly to the national one insofar as fruit trees are concerned, has never been registered; thus, reference data are not available. However, since two increase sections for a total number of 6000 mother plants (which produce and supply 55,000 scions) and at least twenty nurseries are operating in Apulia, an annual production of about 150-180,000 commercial plants may be estimated (O. Murolo, personal communication).

Table II - Citrus productions in Apulia
(ISTAT, 1993)

Variety	Productions
Sweet oranges	
Blond	
Navels	352,562
Common blond	91,350
Valencia	13,745
Ovale o calabrese	12,907
Belladonna	4,941
Blood	
Tarocco	29,202
Sanguinello	10,945
Moro	1,117
Mandarin	
Avana	53,680
Tardivo di Ciaculli	11,245
Clementine (Comune, Oroval, Nules, Fedele etc.)	401,145
Lemon (Ordinario, Femminello ovale e S. Teresa)	37,732

Sanitary status

From the sanitary point of view, Apulian citrus orchards are rather compromised because of the high incidence of viral diseases whose effects reduce the life of the planting with a poor production both in terms of quality and quantity.

The main vector for the spread of these pathologies is undoubtedly the infected propagating material. Moreover the sanitary scenario has been deteriorated by the continuous use of the top grafting technique, which enables to rapidly introduce new citrus varieties of great economic importance in the

existing orchards. Despite the healthy status of old orchards, after a few years most of the grafted varieties show virological symptoms.

So far most of the virus or virus-like diseases have been reported in Italy, among which psorosis is the most spread (Catara,1984). Few foci of tristeza have been found in very restricted areas of Calabria and Sicily, as well as stubborn, whose agent (*Spiroplasma citri)* has never been isolated (Davino and Catara, 1986; Catara *et al.*, 1981).

Due to the lack of data concerning the sanitary status of citrus in Apulia and the relative importance of this crop in the Region, virological investigations have been carried out in the last ten years by the Mediterranean Agronomic Institute in collaboration with the Dipartimento di Protezione delle Piante and the Centro di Studio del CNR sui Virus e le Virosi delle Colture Mediteranee, University of Bari.

A brief account of the results obtained is given in this paper.

Identification of virus and virus-like diseases

Variety collection plots, nurseries and commercial orchards of different citrus varieties and age, with particular reference to the main sweet oranges ('Navel' group), mandarins ('Avana') and clementine ('Common', 'Spinoso', 'Fedele' etc.), were surveyed.

A total of 1750 plants were randomly sampled, in particular 955 in 127 orchards (*c.a.* 2ha in size), 752 in 15 nurseries and 43 in 2 citrus variety collections. Plants showing symptoms of viral origin were also included in the sampling.

Identification of the diseases was done by different detection methods (Roistacher, 1991): (i) visual inspection (in late spring and summer); (ii) biological indexing in woody indicators (virus, virus-like and viroids); (iii) mechanical transmissions to herbaceous hosts (citrus ringspot virus, infectious variegation virus, satsuma dwarf virus, tatterleaf virus); (iv) ELISA (citrus tristeza virus, *Spiroplasma citri*); (v) molecular hybridization (citrus exocortis viroid and cachexia viroid).

- **Virus**

Psorosis (Psorosis A, psorosis B and ringspot) was surely one of the most widespread viral disease in Apulia, especially in the adult citrus groves of susceptible varieties such as sweet orange, mandarin and mandarin-like.

On some local clementines the disease occurred in the form of psorosis-A, which caused a distinct bark scaling of trunks and branches with gummy concavities and wood staining visible in the cross-section of branches. In particular, some groves of the local clementine 'Fedele', that is an early variety of great economic importance, were found infected. Plants were showing symptoms since the tenth year of age and, after 6-8 years, growers thought to overcome the problem by the use of top grafting.

It was not frequently possible to observe leaf mottling in the new flushes because most citrus species were symptomless.

Few reports of ringspot symptoms on mature leaves and /or on fruits were mostly concerning orange varieties such as 'Navelina' in the Ionic area and 'Duretta' in the Gargano area.

Most of the varieties in the groves and in the collection plots proved infected by indexing. A high percentage of infection was detected only in some 'Fedele' local clementine (52%) and 'Navelina' sweet orange orchards (37%). A similar situation was registered in the nurseries, where the local clementine ecotypes and satsuma were the most infected (22% and 25% respectively) whereas sweet oranges, lemons and other citrus showed low rates of infection (8%).

Citrus infectious variegation virus was detected by ELISA and biological indexing in a few plants of sweet orange (2 of 'Valencia', 1 of 'Navelina' and 2 of 'Duretta', the latter being a local orange from Gargano area) and in 8% of lemon groves. Typical crinkly leaf symptoms in association with chlorosis were observed in 1 Valencia and in 2 lemon groves. Very few infected plants (1 orange and 2 lemons) were found in the nurseries by indexing. Only one isolate from 'Duretta' orange was mechanically transmitted to cowpea.

- **Virus-like**

The diseases included in the oakleaf pattern group (concave gum, cristacortis and impietratura) were widely detected in the region. The leaf symptom, frequently observed by indexing and in the new flushes of clementine and 'Navelina' trees in the field, was mainly associated by visual inspections with concave gum, one of the diseases of the group.

Concave gum was the most severe disease in the region affecting all 'Navelina' old clones, which represented the most important orange variety. In this clone severe symptoms were easily observed in the nursery plants and in the very young groves (D'Onghia *et al.*, 1992). 'Nova' hybrid, Satsuma and clementine ('Common', 'Spinoso', 'Precoce di Massafra') were also infected showing mild symptoms.

The most evident symptoms were: production of gum, gum exudation which affects the trunk and the main branches in the mild forms and the twigs in the severe forms. Deep concavities were observed in cross-sections with gumming of annual rings (Figure I). In the most severe cases affecting 'Navelina', wood swelling and cracking occurred with gum exudation since the third year of production.

Many 'Navelina' old clone plantings, showing 70 to 80% of infected plants, were frequently topworked or pulled out for the disease severity.

Impietratura was reported with a low incidence in a few orchards of 'Navelina' sweet orange, lemon and mandarin. In these fruits as hard as a stone, gumming in the albedo corresponding to the discoloured circular spots of the rind was observed.

Cristacortis was not so widespread by visual inspections with the exception of two orchards, where all sour orange rootstock showed typical pitting in a mild and severe form. Symptoms in both rootstock and scion were observed only in four 'Navelina' trees of a single orchard. Moreover mild symptoms in sour orange are often reported in association with the severe concave gum form of 'Navelina' old clone (D'Onghia *et al.*, 1992).

- **Viroids**

Exocortis (CEVd) and cachexia (CCaVd) viroids were widely detected by indexing and molecular hybridization in most of Apulian groves and, slightly less, in the nurseries. Since they are readily mechanically transmissible via working tools, they affect both old and young plantings especially if topworked.

In citrus groves of different species cachexia infection rates were about 82% and slightly less (70%) in the case of exocortis.

Pits in the wood cambium with corresponding bumps and gumming in the bark associated to ***cachexia*** were frequently observed in susceptible species such as tangelo mapo, mandarin and clementine. Infected trees were sometimes stunted and chlorotic, and in some tangelo orchards numerous small pustules on the lower leaf surface were observed.

On the contrary, the typical symptoms of bark cracking and scaling in trifoliate oranges, associated with ***exocortis***, were not observed, since citrus trees were grafted on sour orange rootstock.

Conclusions

With the exception of tristeza virus (CTV), which was detected in one ornamental citrus (*Citrus othaitense*), mainly produced in the Northern Italy, and one lemon of Chinese origin, fortunately indexed before propagation, so far no evidence of quarantine pests has been reported from Apulia. After the first findings of CTV in the surveys carried out in Calabria and Sicily at first and later in Apulia, a national programme for the monitoring of the virus started in 1996, that is still undergoing. Concerning *Spiroplasma citri*, surely Apulia climatic conditions are too cold for the development of this pathogen, that is known to prefer arid and semi-arid climates.

Moreover, a regional project for the improvement of the sanitary status of citrus was presented in 1996 by different Institutions (DPPM-UBA, MAI-B, Centro Ricerca e Sperimentazione "Basile Caramia" of Locorotondo, Istituto Tecnico Agrario of Massafra and Locorotondo). Facilities and research programmes for the production of virus-free propagative budwood were considered in this project with the aim to enhance the citrus productions from the quality and quantitative point of view, particularly referring to the local ecotypes of clementine, which are not included in other international certification programmes.

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