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Serological characterisation of Prunus necrotic ringspot virus (PNRSV) isolates by monoclonal antibodies

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Prunus necrotic ringspot virus (PNRSV), a member of the genus *Ilarvirus*, posses a tripartite genome and morphologically isometric (25-30 nm) to bacilliform shaped particles. The virus was common and widespread in different stone fruits, causing crop losses (Barbara, 1988; Mink, 1992).

PNRSV was phylogenetically related to prune dwarf virus (PDV) and apple mosaic virus (ApMV) (Sánchez-Navarro and Pallás, 1997), and serologic relationship exist between PNRSV and ApMV (Fulton, 1968; Casper, 1973; Halk *et al.*, 1984; Sek-Man Wong and Kenneth Horst, 1993).

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Many isolates of different biological, serological, and molecular characteristics were described and some were sufficiently distinct to be called strains (Barbara *et al.*, 1978; Crosslin and Mink, 1992). The diversity among the isolates was high and formed the bases for differentiating specific biological strains (Mink, 1992).

So far, serological assays have been the most popular method for detecting PNRSV. The first procedure was agar gel double-diffusion test and polyclonal antisera (Fulton and Hamilton, 1960). Mink *et al.* (1987) used to identify three distinct serotypes of PNRSV from sweet cherry trees.

Another serological test, enzyme-linked immunosorbent assay (ELISA) is now the method of choise in the routine detection of PNRSV (Clark *et al.* 1976; Barbara *et al.* 1978; Mink and Aichele, 1984; Uyemoto *et al.*, 1989). Monoclonal antibodies (MAbs) have been produced to several PNRSV isolates (Halk *et al.*, 1984; Jordan *et al.*, 1984).

MAbs to a peach isolate of PNRSV were recently selected and used, at the University of Bari, to investigate the serological properties of the virus. Preliminary tests to characterise the MAbs against 38 PNRSV isolates identified 17 serological variants (Boari *et al.*, 1998).

A large-scale serological detection and characterisation of PNRSV as performed by DASI-ELISA using monoclonal antibodies. PNRSV-positive samples of several stone fruit species from several Mediterranean countries were identified with a MAb cocktail as described by Boari *et al.* (1998). In total, 78 representative PNRSV isolates were collected and characterised against 10 single Mabs. Numbers of PNRSV isolates and locations were: Albania (3), Cyprus (10), Greece (5), Italy (28), Lebanon (6), Malta (5), Tunisia (7), Turkey (7) and Spain (1). In addition, six American isolates were included as comparative controls. The testing of PNRSV isolates, carried out at the Mediterranean Agronomic Institute of Bari, showed high serological variability (33 serogroups among 78 isolates tested), confirming previous reports on serological diversity of PNRSV isolates (Boari *et al.*, 1998). These serogroups were not correlated with host species, geographical origin, or symptom expression.

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