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SEROTYPING OF ALBANIAN *PLUM POX VIRUS* ISOLATES

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SUMMARY - A survey was carried out in Albania to characterise PPV isolates collected from different areas and host species. A total of 48 isolates were identified and typed by DAS-ELISA using monoclonal antibodies (MAbs): 5B (universal), 4DG5 (PPV-D specific), AL (PPV-M specific), AC (PPV-C specific) and EA24 (PPV-EI Amar specific). The majority of Albanian isolates (23) were PPV-M isolates (48%), 12 isolates were PPV-D (25%), whereas 13 of them were natural mixtures of both strains (M+D). Mixed infections were encountered mainly in varietal collections. No isolate was identified as PPV-C and PPV-EA.

Key words: Albania, PPV, virus strain, ELISA, MAbs

RESUME - Une enquête a été conduite en Albanie en vue de caractériser des isolats de PPV provenant de différentes régions et espèces-hôtes. Au total, 48 isolats ont été identifiés et typés par DAS-ELISA, en employant des anticorps monoclonaux (Mabs): le 5B (universel), le 4DG5 (spécifique pour le PPV-D), l'AL (spécifique pour le PPV-M), l'AC (spécifique pour le PPV-C) et l'EA24 (spécifique pour le PPV-EI Amar). La majorité des isolats albanais (23) étaient du type PPV-M (48%), 12 du type PPV-D (25%), alors que 13 étaient des mélanges naturels des deux souches (M+D). Des infections mixtes ont été observées essentiellement dans les collections variétales. Aucun isolat n'a été identifié comme PPV-C et PPV-EA.

Mots-clés: Albanie, souche virale, ELISA, Mabs.

INTRODUCTION

The fruit tree industry is increasing rapidly in Albania considering the drastic reduction at the beginning of the 1990s. With reference to stone fruit trees, native varieties are important particularly for plum and cherry. At present the local nursery activity is reduced and the increasing demand of the domestic market for grafted plants is met mainly by the importation of planting material.

Studies carried out in Albania have shown a compromised sanitary status of stone fruit trees principally due to the presence of sharka disease. The presence of *Plum pox virus* (PPV) is the major threat to the stone fruit industry with a complicated picture composed of PPV-free areas as well as low-rate and high-rate infected regions (Myrta *et al.*, 1994).

PPV isolates fall in four groups which can be differentiated molecularly and serologically: PPV-Marcus (PPV-M), PPV-Dideron (PPV-D), PPV-Cherry (PPV-C) and PPV-EI Amar (PPV-EA). Serological typing by ELISA, giving reliable results (Candresse *et al.*, 1998; Myrta *et al.*, 1998a) is available thanks to monoclonal antibodies (MAbs) with serotype-specific reactivity: PPV-D (Cambra *et al.*, 1994), PPV-M (Boscia *et al.*, 1997), PPV-C (Myrta *et al.*, 2000) and PPV-EA (Myrta *et al.*, 1998c).

First reports on typing a limited number of Albanian PPV isolates were from Myrta *et al.* (1998a,b). In order to complete the previous surveys, a study was extended to different plum-growing areas of the country to characterize Albanian PPV isolates.

MATERIALS AND METHODS

Field surveys and sample collections were concentrated in the main plum orchards in different areas of the country. The following locations were visited: Fushë Mbret (EL), Harizaj (KJ) and Tirana (TR) in

Central Albania; Korçë, Çërravë (KO), Gështenjas, Kuç, Pllaçë (PG), Prrrenjas in Southern-Eastern Albania; Peshkopi, Zdoran (DI) in Northern-Eastern and Shkodër (SH) in Northern areas. The orchards were over 20 years old and a few were varietal collections.

Virus identification was by DAS-ELISA using the universal PPV MAb5B (Agritest, Italy). The serotyping of PPV isolates was carried out with the following MAbs: MAb4DG5 (D-specific), MAbAL (M-specific), MAbTUV and MAbAC (C-specific) and MAbEA24 (EI Amar-specific). In addition, the following MAbs were also used: EA5, EA 8, EA 9, EA 11, EA 12, EA 18 (Myrta *et al.*, 2001) to test for intra-strain serological variability.

RESULTS AND DISCUSSION

The analysis of 48 virus isolates, collected from 12 orchards, showed that 23 (48%) were identified as PPV-M, whereas 13 (27%) as PPV-D (Table 1). Thirteen isolates reacted with both strain-specific MAbs (AL and 4DG5) indicating mixed infection of M and D. No isolate was identified as PPV-C and PPV-EA.

Table 1. Characterization of Albanian isolates of PPV with MAbs

Serotype	Nr. of isolates per strain	Incidence (%)	MAbs reaction				
			5B (univ)	AL (M)	4DG5 (D)	AC (C)	EA 24 (EA)
PPV-M	23	47.9	+	+	-	-	-
PPV-D	12	25.0	+	-	+	-	-
M+D	13	27.1	+	+	+	-	-
Total	48	100					

PPV-M was confirmed as predominant strain in Albania, as previously reported by Myrta *et al.* (1998a,b). It was found in 11 of 12 inspected orchards, confirming its endemic presence in many locations of the country. PPV-D, less distributed than PPV-M, was encountered in 6 orchards coexisting frequently with M strain. Natural mixtures of M and D strain (27% of the total isolates), found in 6 of 12 orchards, were surprisingly higher than expected. In the same orchards, trees with single and mixed infections were found, a situation that occurred more frequent in varietal collections. The majority of tested isolates were originated from plum (Table 2). The analysis of peach and apricot isolates require further investigations.

Table 2. Distribution of PPV strains in different hosts

Host species	Number of isolates		
	PPV-M	PPV-D	M+D
plum	22	11	12
apricot	-	-	1
peach	1	-	-

The serological analysis of Albanian PPV-M isolates with the other MAbs indicated the presence of Mediterranean PPV-M₂ cluster (Myrta *et al.*, 2001). Similar data were collected with Bulgarian and Yugoslavian isolates (data not shown), which support the hypothesis that the Mediterranean PPV-M cluster is the origin foci of PPV in the Balkan peninsula, and subsequent virus evolution in different geographic areas and botanical hosts contributed to the development of current strains.

Considering the high incidence of natural mixtures of strains in Albania and the recent reports of natural recombinant isolates of PPV from plum in Slovakia (Glasa *et al.*, 2001, 2002), it should be of interest to survey for presence of recombinant isolates under Albanian conditions.



Fig. 1. Distribution map of PPV strains in Albania

This work reports the status of PPV strains in Albania as surveyed orchards were old. In the last decade, many small-sized peach and apricot orchards have been established in the coastal area of the country with propagating material frequently coming from abroad. More investigations in these new fruit tree growing areas are needed.

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