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SEROLOGICAL CHARACTERIZATION OF MOROCCAN SOURCES OF CITRUS PSOROSIS VIRUS (CPsV)

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SUMMARY - CPsV detection by ELISA and biological indexing was carried out on 4 Moroccan sources, used as psorosis positive controls at UCP, Rabat. Two sources were ELISA-positive, showing psorosis-like symptoms whereas for the remaining one, it was not a true psorosis and one was apparently not serologically detected.

Key words: Citrus, citrus psorosis virus, ELISA, Morocco

RESUME - *La détection par ELISA et indexage biologique a été réalisée sur 4 sources Marocaines, utilisées comme contrôle positif à l'UCP, Rabat.*
Deux sources ont réagi positivement au test ELISA, montrant des symptômes associés à la psorose. Il s'est avéré qu'une source n'était pas une vraie psorose, par contre la dernière source semble ne pas être détectée sérologiquement.

Mots clés*: Agrumes, virus de la psorose des agrumes, ELISA, Maroc*

INTRODUCTION

Several graft-transmissible pathogens are known to affect the Moroccan citrus industry (Bové, 1995) among which citrus psorosis virus (CPsV) is considered one of the most widespread (Nhami and Zidane, 1984). This virus may induce severe bark scaling symptoms in the trees or may be symptomless in most of citrus species.

Protocols for serological detection of CPsV by ELISA have recently been developed (Garcia *et al*., 1997; D'Onghia *et al*., 1998; Alioto *et al*., 1999; Potere *et al*., 1999; Djelouah *et al*., 2000; Roistacher *et al*., 2000) the use of which highly improved virus identification and characterization. Specific CPsV antibodies can easily separate psorosis from other diseases (concave gum, cristacortis, impietratura, infectious variegation) responsible for leaf discolorations and oak leaf patterns symptoms.

Based on these findings a study was conducted in Morocco for the serological characterization of Moroccan citrus accessions, used as positive controls at Direction des Domaines Agricoles, Unité de Contrôle des Plants (UCP) - Rabat, Morocco.

MATERIALS AND METHODS

The 20-year-old Moroccan accessions of 3 sweet oranges (1 Valencia late, 2 Washington navel) and 1 Shambar grapefruit, used as psorosis sources at UCP, were selected and referred as Ps-M 1, M2, M3, M4. All of them were indexed on Madame vinous sweet orange, Dweet tangor and Carte Noir mandarin (Roistacher, 1991).

Monoclonal antibodies (Mabs Ps), provided by Agritest Company in Italy, were used following the procedure of Potere *et al*. (1999). Samples were collected from young and mature leaves, symptomatic and symptomless. Sampling was carried out during Spring in the field and in the greenhouse. The homologous Italian IAMB Ps 191X and the healthy tree were included in the tests as positive and negative controls.

The Mabs Ps cocktail diluted 1:500 in carbonate buffer pH 9,6 was used for plate coating and the alkaline phosphatase conjugated-Mab diluted 1:1000 in PBS was used for antigen detection. Samples were considered positives if their values were over 0,1 OD and 2 times or more higher than healthy citrus plant extracts used as control.

RESULTS AND DISCUSSION

Results of indexing correlated with those of ELISA (Tab.1). Ps-M3 and Ps-M4 were ELISA-positive and showed shock symptoms (Madame vinous and Carte Noir mandarin) and mottling in the leaves of the indicators. No serological reaction was obtained with Ps-M1 and Ps-M2, which showed only oak leaf patterns in the emerging leaves of Madame vinous and Carte Noir. These symptoms are associated to another group of diseases (concave gums, cristacortis and impietratura), thus confirming ELISA results.

No differences were noticed in the results where symptomatic leaves were used. Moreover, mature leaves showed higher absorbance values compared to the young ones.

CONCLUSION

The use of ELISA technique was highly successful for the assessment of true psorosis sources, with important effect in pathogen detection in the framework of certification. It successfully separated psorosis from a complex of diseases which induce oak-leaf pattern symptoms in young leaves. The combination of ELISA with biological indexing can highly improve virus detection for the production of a primary source in the framework of certification. Moreover, this technique permits to carry out CPsV detection on a large scale for acquiring useful information on the presence, incidence and distribution of the virus in a certain area. A further study with a panel of Mabs Ps raised against the Italian CPsV isolate (Potere *et al.,* 1999; Djelouah *et al*., 2000) and the two Mabs Ps raised against an American virus isolate (Alioto *et al*., 1999) is desirable in order to complete the serological characterization of the tested Moroccan CPsV sources by serological means.

Table 1. Results of psorosis detection by DAS-ELISA and biological indexing.

Sources	Varieties	**ELISA**	**Biological indexing**		
			D.T.	**M.V.**	**C.N.M.**
Healthy	Washington navel	-	-	-	-
Ps- M1	Valencia late	-	OLP	-	-
Ps- M2	Washington navel	-	OLP	OLP	OLP
Ps- M3	Shambar	+	Mot, Sh	Mot, Sh	Mot
Ps- M4	Washington navel	+	Mot, Sh	Mot, Sh	Mot

C.N.M.: Carte Noire mandarin; D.T.: Dweet tangor; M.V.: Madame vinous orange; OLP: oak leaf patterns;

Sh: leaf shock; Mot: leaf mottling n

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