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Sanitary status of Spanish autochthonous almond cultivars and its implication in breeding strategies

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Abstract. Traditional almond cultivars are an interesting source of different traits for breeders. These old cultivars have been multiplied by grafting for a long time and usually have been accumulating different viruses. The objective of this work is to determine the presence of different virus in these cultivars by multiplex RT-PCR. Virus analysed included: *Apple chlorotic leaf spot virus* (ACLSV), *Plum pox virus* (PPV), *Apple mosaic virus* (APMV), *Prunus necrotic ring spot virus* (PNRSV), *Prune dwarf virus* (PDV), *Apricot latent virus* (APLV), *Plum bark necrosis and stem pitting-associated virus* (PBNSPaV), and *American plum line pattern virus* (APLPV). The obtained results showed the generalized presence of PNRSV and PDV in the analyzed collections with a preesence of 90% and 68% respectively. Despite of these virus do not show noticeable symptoms in the tree nor fruits, these poor phytosanitary state urge to the adoption of new estrategies for elimination of virus and cleaning these materials.

Keywords. Almond – Germplasm – Virus – RT-PCR – Sanitary status.

Statut sanitaire des collections espagnoles autochtones d'amandier et son implication dans les stratégies de sélection

Résumé. *Les cultivars d'amandier traditionnels sont une source intéressante de différents traits pour les éleveurs. Ces anciens cultivars ont été multipliés par greffage depuis longtemps et généralement ils ont accumulé virus différents. L'objectif de ce travail est de déterminer la présence du virus dans ces différents cultivars par RT-PCR multiplex. Les virus analysés on été: Apple chlorotic leaf spot virus (ACLSV), Plum pox virus (PPV), Apple mosaic virus (APMV), Prunus necrotic ring spot virus (PNRSV), Prune dwarf virus (PDV), Apricot latent virus (APLV), Plum bark necrosis and stem pitting-associated virus (PBNSPaV), et American plum line pattern virus (APLPV). Les résultats obtenus montrent la présence généralisée de PDV et PNRSV dans les collections analysés avec un présence de 90% et 68% respectivement. Malgré que ces virus ne présentent pas de symptômes visibles dans l'arbre, ni dans les fruits, le pauvre état phytosanitaire exhorte à l'adoption de nouvelles stratégies pour l'élimination de virus et de nettoyage de ces matériaux.*

Mots-clés. *Amandier – Germoplasme – Virus – RT-PCR – SSR.*

I – Introduction

Traditional almond cultivars are an interesting source of different traits for breeders. These old cultivars have been multiplied by grafting for a long time and usually have been accumulating different viruses.

Although no data is available in almond, *Prunus* orchards are affected by numerous viruses including *Plum pox virus* (PPV), causing sharka disease, and *Apple chlorotic leaf spot virus* (ACLSV). These are the most widespread viruses, followed by ilarviruses like *Prunus necrotic ring spot virus*

(PNRSV), *Apple mosaic virus* (ApMV) and *Prune dwarf virus* (PDV) (Dominguez *et al*., 1998; Myrta *et al*., 2003; García-Ibarra *et al*., 2012). The following are other new viruses described in Spain: *Plum bark necrosis and stem pitting-associated virus* (PBNSPaV), *American plum line pattern virus* (APLPV) and *Apricot latent virus* (APLV) (García-Ibarra *et al*., 2010).

The objective of this work was to determine the presence of different viruses in the Spanish autochthonous almond collections from the breeding programs of CEBAS-CSIC of Murcia and IRTA of Reus by multiplex RT-PCR. Virus analysed included ACLSV, PPV, APMV, PNRSV, PDV, APLV, PBNSPaV and APLPV.

II – Materials and methods

Almond genotypes assayed included 44 early flowering and self-incompatible local cultivars from the almond collections of CEBAS-CSIC of Murcia and IRTA of Reus (Table 1).

Leaf samples were analysed by multiplex RT-PCR to detect eight viruses, including *American plum line pattern virus* (APLPV), *Apple chlorotic leaf spot virus* (ACLSV), *Apple mosaic virus* (ApMV), *Apricot latent virus* (ApLV), *Plum bark necrosis and stem pitting-associated virus* (PBNSPaV), *Prune dwarf virus* (PDV), *Prunus necrotic ringspot virus* (PNRSV) and *Plum pox virus* (PPV) (Sánchez-Navarro *et al*., 2005).

III – Results and discussion

The results of this work show the high rate of infections in the Spanish autochthonous almond collections from CEBAS-CSIC of Murcia and IRTA of Reus. These results showed the generalized presence of PNRSV and PDV in the analyzed collections with a preesence of 90% and 68%, respectively. However no presence of ACLSV, PPV, APMV, APLV, PBNSPaV or APLPV was detected (Table 1).

These levels of infection are much higher than previously detected by serological techniques in almond (48% infection with PNRSV and PDV 23%) (Myrta *et al*., 2003). PNRSV is distributed worldwide in *Rosa* and *Prunus* spp. Generally, symptoms of PNRSV appear in the first year after infection, showing as asymptomatic in subsequent years, except for some isolates causing recurrent symptoms each year. The second multiplex RT-PCR carried out on CEBAS-CSIC collections showed the same results.

The potential yield loss caused by PNRSV in almond is usually low. PDV is similar to PNRSV in worldwide distribution. It is one of the most important viruses of stone fruit trees, predominantly sour and sweet cherry. PDV causes yellowing of leaves in these species and the dwarf peach alone or in mixed infections with PNRSV. In almonds were not described symptoms. Although these viruses are not a major threat to the almond tree, this species may play an important role in the spread of these viruses to other fruit, its control in nurseries, plantations and genebanks still needed.

IV – Conclusions

The obtained results showed the generalized presence of PNRSV and PDV in the analyzed collections with a presence of 90% and 68%, respectively. Despite the fact that these viruses do not show noticeable symptoms on the tree or fruits, this poor phytosanitary state calls for the urgent adoption of new strategies for the eliminating the viruses and cleaning up these materials.

Table 1. Spanish autochthonous almond cultivars assayed from the CEBAS-CSIC of Murcia and the IRTA of Reus collections

		Virus							
Variety	**Origin**	**ACLSV**	**ApLV**	**APLPV**	**ApMV**	**PBNSPaV**	**PDV**	**PNRSV**	**PPV**
CEBAS									
Atascada	Murcia	–	–	–	–	–	+	+	–
Atocha	Murcia	–	–	–	–	–	–	+	–
Avellanera	Murcia	–	–	–	–	–	+	+	–
Bonita	Baleares	–	–	–	–	–	+	+	–
Carretas	Murcia	–	–	–	–	–	+	+	–
CEBAS–1	Murcia	–	–	–	–	–	+	+	–
Colorada	Murcia	–	–	–	–	–	–	+	–
Del Cid	Desconocido	–	–	–	–	–	+	+	–
Desmayo AD	Incierto	–	–	–	–	–	+	+	–
Desmayo Lorca	Murcia	–	–	–	–	–	+	+	–
Fina del Alto	Murcia	–	–	–	–	–	–	+	–
Fournat	Francia	–	–	–	–	–	–	+	–
Garrigues	Murcia	–	–	–	–	–	+	+	–
J. Salazar	Murcia	–	–	–	–	–	+	+	–
Jordi	Mallorca	–	–	–	–	–	–	+	–
La Mona	Murcia	–	–	–	–	–	+	+	–
Malagueña	Desconocido	–	–	–	–	–	–	+	–
Marcona	Alicante	–	–	–	–	–	+	+	–
Marcona AD	Murcia	–	–	–	–	–	–	+	–
Marcona de San Joy	Murcia	–	–	–	–	–	+	+	–
Marcona Flota	Murcia	–	–	–	–	–	+	+	–
Pajarera	Murcia	–	–	–	–	–	+	+	–
Peraleja	Murcia	–	–	–	–	–	+	+	–
Planeta Fina	Alicante	–	–	–	–	–	+	+	–
Planeta Roja	Alicante	–	–	–	–	–	–	+	–
Ramillete	Murcia	–	–	–	–	–	–	+	–
Rumbeta	Alicante	–	–	–	–	–	+	+	–
Verruga	Murcia	–	–	–	–	–	+	+	–
IRTA									
Angones	Lleida	–	–	–	–	–	+	+	–
Asperilla	Huesca	–	–	–	–	–	+	+	–
Belardino	Castellón	–	–	–	–	–	+	–	–
Caima	Lleida	–	–	–	–	–	–	+	–
Carreró	Castellón	–	–	–	–	–	+	+	–
Esperanza forta	Tarragona	–	–	–	–	–	+	–	–
Gabaix	Tarragona	–	–	–	–	–	+	+	–
Mollar de la Princesa	Francia	–	–	–	–	–	+	–	–
Mollar de Tarragona	Tarragona	–	–	–	–	–	+	+	–
Nano	Castellón	–	–	–	–	–	+	+	–
Parque Samà	Tarragona	–	–	–	–	–	+	–	–
Pauet	Lleida	–	–	–	–	–	–	+	–
Pep de Juneda	Lleida	–	–	–	–	–	+	+	–
Rof	Tarragona	–	–	–	–	–	–	+	–
Tardaneta	Tarragona	–	–	–	–	–	–	+	–
Verd	Castellón	–	–	–	–	–	+	–	–

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