

BULGARIAN POTATO CULTIVARS SENSITIVE TO POTATO VIRUS Y IN FIELD AND GREENHOUSE CONDITIONS

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ABSTRACT

Plant viruses are a major threat to seed production and potato yield in Bulgaria. The main plant viruses infecting potatoes are Potato virus Y (PVY), Potato leaf roll virus (PLRV), Potato virus A (PVA), Potato virus X (PVX), Potato virus V (PVV), Tobacco rattle virus (TRV) and Tomato spotted wilt virus (TSWV), of which PVY and PLRV are of greater economic importance. Bulgarian seed potato cultivars grown in the field and in green house conditions were tested for the presence of viral infection. Many cultivars grown in the field were carriers of viral infection. Major roles in the spread of viruses have altitude, geography of the planting region, use of healthy planting material and agricultural activities. The determination of the sensitivity of the Bulgarian potato cultivars to PVY will help to control the seed material which is essential for obtaining high quality production.

Key words: PVY, potatoes, resistance

Introduction

Potatoes originated in the Peruvian Andes and belong to the *Solanaceae* family. The commonly cultivated potato (*Solanum tuberosum*) belongs to the series *Tuberosa*, one of 19 series of the subsection *Potatoe*, which comprises 225 wild tuber-bearing species (Hawkes, 1992). Potato was introduced into Europe around 1570 into Spain (Hawkes, 1992). Nowadays the potato is one of the world's major crops with an annual production of up to 300 million tons in 2003. According to recent FAO statistics (FAOSTAT, 2003), the potato ranks 5 in worldwide food production after sugar cane, maize, rice and wheat. Europe is the largest potato producer in the world producing 130 million tons (42.1 %). In potato, selection is performed by evaluation of more than 50 traits with respect to different utilization purposes and it takes more than 15 years to generate a new cultivar.

Potato virus Y (PVY) is one of the most important viruses in *Solanaceae* (tomato, tobacco, pepper) and can reduce potato production up to 80% (De Bokx & Huttinga, 1981). PVY is also wide spread and in some parts of Europe it is more common than the Potato Leaf Roll Virus (PLRV). PVY caused severe damage in potato seed production. A new strain (PVY^{NTN}) causing big necrotic rings on the tubers is presently attacking potatoes and affects also the ware potato market (Weidemann, 1993).

There are two resistance mechanisms in plant cells against invading viruses. The first resistance mechanism can be characterized by the lacking of infection (immunity) or by strong reduction of virus replication in infected cells. The second resistance mechanism inhibits virus spread from cell-to-cell and through the vascular system (Valkonen & Somersalo, 1996; Bendahmane *et al.*, 1999). Two major types of mono-genetically inherited resistance genes to PVY are known in cultivated and wild potato species (*Solanum* spp.), namely hypersensitive resistance (*Ny*) and extreme resistance (*Ry*) also designated as immunity (Ross, 1986). The hypersensitivity reaction inhibits virus movement and belongs to the second group of resistance but does not provide a total defense against PVY. Immunity or extreme resistance to PVY (*Ry*) was first reported by Cockerham (1943) in *Solanum stoloniferum*, which is a Mexican wild potato species, which belongs

to *Longipedicellata* Buk. (Cockerham, 1970; Hawkes, 1992). No PVY could be detected with ELISA in inoculated plants expressing *Ry* (Ross, 1986). Ross described approximately 20 cultivars carrying the gene *Ry* for extreme resistance to PVY derived from *S. stoloniferum*. Potatoes cultivars Bettina, Forelle and Ute were registered as cultivars with PVY-immunity. Potato cultivars Kuras, Sibü, Petra and Tomba were classified as highly resistant to potato virus Y while cultivars Laura and Sempra were recorded as susceptible to PVY.

In Bulgaria the situation with resistance of potato cultivars to PVY in the field conditions is still unknown and the aim of this research is to determine the resistant and the sensitive to PVY infection potato cultivars.

Material and methods

Plant material: Potato cultivars from Maritsa Vegetable Crops Research Institute: Nadejda, Pavelsko, Iverce, Orfey, Bor, Perun, Rojen, Kalina.

DAS-ELISA (Double Antibody Sandwich Enzyme Linked Immunosorbent Assay):

The analysis was conducted by the method of Clark and Adams (1977). We have used a commercial kit of LOEWE Biochemica GmbH, Sauerlach, Germany. ELISA plates are loaded with antiserum (IgG) for PVY, with dilutions (according to the instructions of the manufacturer) in 0.05 M carbonate buffer. The samples were incubated for 4 hours at 37 ° C, and the unbound components were washed out with PBS-T buffer for 5 min. All samples were grounded in extraction buffer containing 1% PVP (polyvinyl pyrrolidone) in a ratio of 1:10. The plates were incubated at 4 ° C for 16 hours. Following the third wash step alkaline-phosphatase conjugate for PVY was added and the plates were incubated for 4 hours at 37 ° C. The used substrate is p-nitrophenyl phosphate (p-nitrophenyl phosphate, Sigma) in diethanolamine buffer (pH 9.8) at a ratio of 1mg/ml. The reaction proceeded in the light at room temperature and was stopped with 3N NaOH. The adsorption of the color reaction is measured at multifunctional detector (DTX 880) at a wavelength of 405nm.

The positive samples had optical density (OD) over the threshold (Cut-off) which is three times the value of the negative control.

Results and discussion

We use germs from potato seed tubers from all eight Bulgarian potato cultivars as a plant material for DAS ELISA assay. The seed tubers from all the cultivars tested were symptomless. In spite this in the potato cultivar Rojen PVY was present (Fig.1, Fig.2).



Fig. 1 Potato tuber from cv. Rojen without symptoms but infected with PVY

In other potato cultivars such as Agria, Arinda, Marabel, grown in different geographic regions in Bulgaria, PVY is present in some potato tubers according to the production region. In Samokov, Bansko and Velingrad, regions with high altitude and mountain climate PVY is not

present. In contrast in other regions like Pernik, Trun and Pazardjik PVY is present in these potato cultivars.

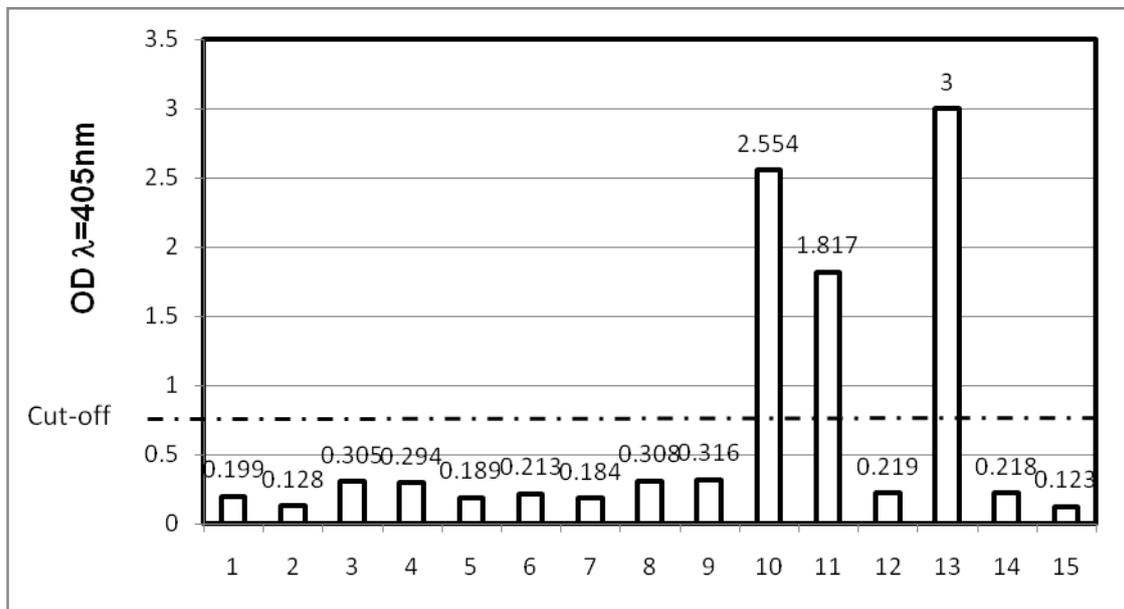


Fig.2 DAS-ELISA results for PVY infection in Bulgarian potato cultivars

Legend: 1 – potato cv. Nadejda in the field conditions; 2 - potato cv. Nadejda in the green house conditions; 3- potato cv. Pavelsko in the field conditions; 4 - potato cv. Iverce in the field conditions; 5 - potato cv. Orfey in the field conditions; 6 - potato cv. Orfey in the green house conditions ; 7 - potato cv. Bor in the field conditions; 8 - potato cv. Perun in the field conditions; 9 - potato cv. Perun in the green house conditions; 10 - potato cv. Rojen in the field conditions; 11 - potato cv. Rojen in the green house conditions; 12 - potato cv. Kalina in the green house conditions; 13 – K+ for PVY from the kit; 14 – K- for PVY from the kit; 15 – Buffer

It was interesting that potato cultivar Rojen was infected with PVY both in the field and in green house conditions (Fig.2). All the other tested potato cultivars were virus free (Fig.2).

These results lead to the conclusion that most probably PVY was spread in cv. Rojen through infected tubers and not the other way of virus transmission.

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