

D SHARKA

D1 Varietal and clonal differences in a rate of infection by PPV within *Prunus domestica* L.

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Since 1998 the spread of plum pox virus (PPV) was monitored in several experimental and commercial plum orchards established in the Czech Republic. This study was focussed on differences in this spread between cultivars and also between different clones within 'Domestic Prune'. A majority of cultivars were evaluated in an experimental orchard established in 1993. Nearly one thousand of the trees were grown in plots located closely to germplasm plum orchard where about 80 % of trees were infected by plum pox virus. All newly infected trees were immediately discarded after identification of their infection by PPV. The highest shares of infected trees were noted with 'Empress' cv, followed by 'President', 'Early Blue', 'Valor' and 'Wegierka Dabrowicka'. On the other hand no infected tree was found with 'Nancy Mirabelle'. Low shares of infected trees were observed with 'Valjevka', 'Čačanska rana', 'Reine Claude', 'Hamanova' and 'Bluefre'. In the case of 'Čačanska lepotica', 'Čačanska najbolja', 'Domestic Prune', 'Stanley' and some other cultivars trees were infected by intermediary shares between the groups.

A somewhat different pattern of cultivar infection took place in two other experimental orchards without removing of infected trees. There, the highest shares of infected trees were observed with 'Domestic Prune' cv. and some other cultivars closely related to it. On the other hand all trees of 'Opal', 'Oullins' and 'Reine Claude' remained without any infection. Very low rates of the infection were found in 'Bonne de Bry', 'Nancy Mirabelle', 'Pamjat Vavilova', 'Wazons Reine Claude' and 'Wengerka Jubilejnaja'. In commercial orchards growing under low infection pressure of PPV from surroundings, trees of 'Gabrowska' cv. were less infected than those of 'Čačanska lepotica', 'Čačanska najbolja', 'Domestic Prune' or 'Stanley'. Significant differences in the rate of PPV infection were also observed between clones within 'Domestic Prune' cultivar.

D2 The occurrence of plum pox virus in Norway

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In 1998 PPV was detected for the first time in Norway. Virus symptoms were observed on several trees in a collection of plum cultivars at Njøs Research Station in Sogn og Fjordane County in the western part of Norway.

In 1998 all collections of plum varieties and nuclear stock collections of plum in Norway were tested. All fruit tree nurseries were inspected, and samples were collected for ELISA-testing. The main nuclear stock collection of plum at Sauherad Elite Plant Station in Telemark County in southeast of Norway, had no PPV infection, whereas a small nuclear stock orchard at Njøs turned out to have infected trees of the plum cultivars 'Mallard' and 'Rivers Early Prolific'. Unfortunately some fruit

growers and nurseries, especially in the western part of Norway, have received infected material from this source.

Since 1998 most commercial plum orchards in Norway have been surveyed. About 60 000 individual trees have been tested. About 1 % of the trees have been found infected by PPV. We believe the main infection source must have been infected plums or apricots imported to Njøs around 1970 or earlier.

It has been some spread of PPV by aphids in certain areas, but on the whole, the spread by aphids is considered to be absent or slow in Norwegian plum orchards, and we expect that PPV can be eradicated from commercial plum orchards in Norway.

D3 A strategy to bring under control the losses of Sharka in Bulgaria

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Since 1932 when sharka was identified for the first time in Bulgaria by Prof. D. Atanasov up to the present days this disease spread in the many regions of the country. During the 70^s the very susceptible local cultivar Kyustendilska plum was replaced by cv. Stanley and thus the plum production was temporarily restored, but on the other hand, the utilization of only one plum cultivar in the orchards causes additional production and market problems.

In connection with the epidemiological situation and the heavy losses suffered researchers have increased their efforts in recent years in order to elaborate a strategy for control of sharka damages in Bulgaria. This strategy includes the following key elements:

- Introduction of certification scheme for the production of virus free planting material of plum;
- Enrichment of the national list of plum cultivars by introduction of foreign cultivars and conducting of own breeding for sharka resistant and tolerant cultivars;
- Developing of new intensive technologies for plum production that will ensure control of sharka spreads and fast return of investments.

D4 70 years investigation on Sharka in Bulgaria

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Prof. Atanasov has published for first time a paper "Plum pox . A new virus disease " during 1933. He starts this investigation in 1926 at the region of South-West Bulgaria where on Kyustendilska sinya plum have been observed unknown disease since period about World War I. During the summer of 1931 after budding on seedlings of *P.cerasifera* he proved the virus character of this new disease.

In the " Diseases on cultivated plants" book Atanasov (1934) described the symptoms on plum, sweet cherry, peach and apricot cultivars with photos of symptoms on leaves and fruits. Since those time for control of the sharka he recommended to eradicate all diseased trees from the orchards and nurseries and to stoped export of budwoods and plants from regions of sharka. The investigation on the sharka in our country we can separate in three stages:

- First stage (1926 -1970) when the symptoms on plums, peaches, apricots, cherries and almond have been described and the host plant are also investigated. Visual and biological test by *C. foetidum* indicator are the major diagnostic methods;
- During second stage (80^s) by ELISA test has been investigated lot of cultivars, hybrids and rootstocks;
- Third stage (after 1990) identification of PPV stain by serological and molecular methods.

The host plants as weeds are investigated also and by in vitro techniques are produced virus free planting material. The main goal in the breeding programme is tolerant and resistant stone fruit cultivars.

D5 Evaluation of PPV on Plum Fruits

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Genebank of plums on myrobalan rootstocks (6 x 4 m spacing and two trees per cultivar) was planted in 1990. In 1991, 207 plum cultivars were artificially inoculated by Plum Pox Virus (PPV) on site. The source of PPV (M strain) was taken from the tree of domestic plum, which was free of other viruses. Three buds infected with PPV were budded on one-year-old trees. In 1996, trees with negative results in ELISA were reinfected with the same isolate of PPV. Since 2001, incidence of PPV is evaluated on plum fruits according to descriptor.

- 1 – incidence of deformations of fruit surface (very strong, up to 100 % surface of fruits damaged).
- 3 - incidence of deformations of fruit surface – medium to strong, up to 50 % surface of fruits damaged
- 5 - incidence of deformations of fruit surface – low, up to 10 % surface of fruits damaged
- 7 –color changes on skin only, or sporadic incidence of small deformations
- 9 – fruits without symptoms

Symptoms of PPV on fruits were not found in 44 cultivars of plums. For example, following cultivars belong to this group: Stanley, Opal, Oullins Gage, Cacanska najbolja, Cacanska leptica, Gabrovska, Green Gage, Tuleu Timpuriu. On the other hand, 29 cultivars were included in group 1 (for example all clones of Hauszwetsche, Bucurie). This work was supported by the Czech Ministry of Agriculture (Project NAZV- QD 1407)

D6 Variability of resistance to Plum Pox Virus in individual naturally growed trees of Myrobalan

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Myrobalan trees (*Prunus cerasifera* L., ssp. *myrobalana*) are often naturally growed in lowland and hill regions of Bohemian and Moravian parts of the Czech Republic (CZ) at 200-400 meters above sea level. The occurrence of natural infection of myrobalan trees with Plum pox virus (PPV) was proved already in 1989. Recently, myrobalan was proved to be second main natural source of PPV infection in the CZ. On the other hand many of myrobalan trees are rather resistant to the PPV.

Trees of myrobalan infected with PPV were evaluated for the resistance to plum pox. The intensity of PPV symptoms in leaves and fruits was inspected and relative concentration of PPV protein in leaves was determined by ELISA.

Leaves of most PPV infected myrobalans carry only very mild mosaic in leaves and mild diffuse spots in fruits, some trees are symptomless. Occasionally medium severe, or severe symptoms were observed in leaves and fruits. The relative concentration of PPV protein in leaves of individual trees fluctuated from 5.0×10^{-2} to 1.56×10^{-3} . Myrobalans are more resistant to PPV in comparison with plums or blackthorns. There is a high probability to obtain clones of myrobalan, as a natural source of resistance to PPV.

D7 Leaf and fruit susceptibility of European plum cultivars to plum pox potyvirus in Hungary

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Leaf and fruit susceptibility of 25 commonly grown European plum cultivars to plum pox virus were evaluated in five not consecutive years, during the period of 1988-2003. The evaluated cultivars were 'Albatross', 'Althanova ringlota', 'Asatan', 'Bartschis Frühzwetsche', 'Besztercei szilva', 'Bluefre', 'Bourdett Angelina', 'Čačanska leptica', 'Čačanska najbolja', 'Čačanska rana', 'Čačanska rodna', 'California Blue', 'Chrudimer', 'Debreceni muskotály', 'Early Italian', 'Fellenberg T.24', 'Frühe Fellenberg', 'Hauszwetsche Grider', 'Hauszwetsche Rudin', 'Kisinyevszkaja Rannaja', 'Myrabelle de Nancy', 'Ontario', 'Pescarus', 'Richards Early Italian' and 'Silvia'. The trees were planted at different times from 1978 to 1982 and grafted on *P. cerasifera* rootstock. Leaf and fruit incidence as well as severity were assessed according to a grade ranging from 0 to 5. Assessments were made at harvest time of each cultivar on 100 leaves and 100 fruits per tree in four replicates. After the five-year assessment, cultivars were classified into 5 groups (highly susceptible, susceptible, moderately susceptible, low susceptible, not susceptible) according to their susceptibility to plum pox virus. For both leaf measurements, mean incidence and severity of plum pox infections were the highest on cv. 'Albatross' and the clones of cv. 'Besztercei szilva' and the lowest on cv. 'Ontario'. Fruit symptoms were the most severe on cv. 'Albatross' and the clones of cv. 'Besztercei szilva' and no fruit symptoms were found on cv. 'Čačanska rana', 'Myrabelle de Nancy' and 'Ontario'.

D8 Updated characterization of plum pox virus isolated from the balkanic area to Western Europe

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We have recently collected diverse isolates of plum pox potyvirus (PPV) known to infect *Prunus domestica*. The data reported in this study confirm that the domestication of these isolates in greenhouse conditions led to the appearance of PPV variance. This indicates that the passage to herbaceous hosts can provide important insights into biological properties.

From the phytopathological studies under confined greenhouse conditions, we have made the immunological and molecular analyses of these PPV isolates. Some differences in PPV capsid cistron enabled us to confirm the diversity of the *Prunus* species sources that is thought to be the main cause. Given these molecular data, the investigation on the 5' part of PPV genome is underway. Alignment of nucleotide sequences will be made and the significance of these results will be discussed.