

# STUDY REGARDING THE EVALUATION OF THE ATTACK DEGREE OF THE PLUM POX VIRUS (PPV) AT SOME PLUM CULTIVARS (*PRUNUS DOMESTICA* L.)

## STUDIUL PRIVIND EVALUAREA GRADULUI DE ATAC AL VIRUSULUI PLUM POX (PPV) LA UNELE SOIURI DE PRUN (*PRUNUS DOMESTICA* L.)

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**Abstract.** Between the years 2008 and 2009, at Fruit Growing Research & Extension Valcea, were evaluated twenty plum cultivars regarding the attack degree of the Plum pox virus. Performing the serologic test Double Antibody Sandwich-Enzyme Linked Immunosorbent Assay was established that only six were not infected with this virus: Tuleu timpuriu, Diana, Flora, Minerva and Vangerka jubilejnija. On leaves, the different values of the frequency of the attack oscillated between 12% and 94,5%, and of the intensity between 21,2% and 61,5%. The limits between the attack degree oscillated were 2,5% and 55,4%. Regarding the virus attack on fruits, only five cultivars were affected: Renclod hramova, Kisinevskaia rana, Valor, Balada and Edda.

**Key words:** frequency, intensity, concentration of the viral protein, ELISA.

**Rezumat.** În perioada 2008-2009, la Stațiunea de Cercetare-Dezvoltare pentru Pomicultură Vâlcea, au fost evaluate douăzeci de soiuri de prun privind gradul de atac al virusului Plum pox. În urma efectuării testului serologic Double Antibody Sandwich-Enzyme Linked Immunosorbent Assay s-a stabilit că doar șase sunt libere de virus, respectiv: Tita, Tuleu timpuriu, Diana, Flora, Minerva și Vangerka jubilejnija. Pe frunze, frecvența atacului s-a manifestat diferit, valorile oscilând între 12% și 94,5%, intensitatea atacului a oscilat între 21,2% și 61,5%, iar pentru gradul de atac s-au înregistrat valori cuprinse 2,5% și 55,4%. Pe fructe, atacul virusului a fost înregistrat doar la 5 soiuri: Renclod hramova, Kisinevskaia rana, Valor, Balada și Edda.

**Cuvinte cheie:** frecvență, intensitate, concentrația proteinei virale, ELISA.

## INTRODUCTION

Plum pox virus (PPV) is a Potyvirus that infects many *Prunus* ssp. The virus causes a disease called Sharka which is one of the most devastating diseases of stone fruit industry in Europe and the Mediterranean (Roy and Smith,1994). The disease is detrimental primarily for apricot, peach, and plum trees, reducing quality

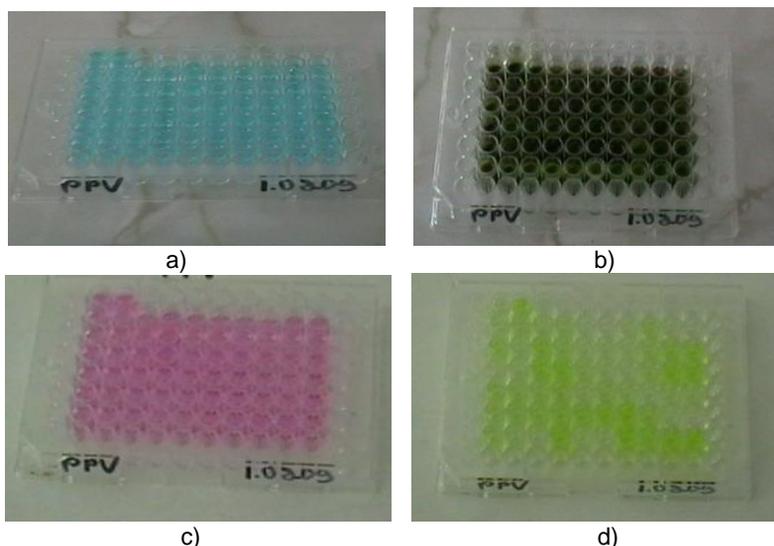
and causing premature dropping of fruits (Dunez and Sutic, 1988). The severity of symptoms is influenced by host species and cultivar and, to a lesser degree, by climate, nutrition and growth of the trees, their age, the viral strain etc. (Desvignes,1999).

The aims of present study were: the identification of the virus in the sap of several stone fruit samples and to determine the attack degree on leaves and fruits in case of the cultivars infected.

## MATERIAL AND METHOD

The biological material was represented by twenty plum cultivars (*Prunus domestica* L.): Tita, Stanley, Renclod hramova, Tuleu timpuriu, Anna Spath, Kisinevskaja rana, Large sugar prune, Diana, Robé de Sergent, Verzișoare de Olteanca, Imperial California, Čačanska rodna, Flora, Edda, Tuleu gras de Sinești, Vangerka jubilejnija, Minerva, Valor, Balada and Tegera.

The concentration of the viral protein in leaves was determined through the serologic method Double Antibody Sandwich-Enzyme Linked Immunosorbent Assay (DAS-ELISA) (Clark&Adams,1977). The PPV reagents were purchased from the firm Bioreba (Switzerland) and the work protocol was done according to the technical information received in the kits brochure (Figure 1).



**Fig. 1.** ELISA Microplates: - **a.** The distribution of a specific antibody; **b.** The distribution of the plant extract; **c.** The distribution of the conjugate; **d.** The distribution of the substrate with pNPP: color reaction indicates infected sample.

The attack degree (AD%) was calculated according to the formula:  $AD\% = (F\% \times I\%) / 100$  (Cociu, 1989) and it represents the expression of the suffering infected plants for the years in questions.

The frequency of the attack was calculated using the formula:  $F\% = (n \times N) / 100$ , in which "n" means the number of the affected plant organs referred to the total number of analysed plant organs "N".

The intensity of the attack was determined applying the formula:  $I\% = \sum(i \times f) / b$ , in which "i" means the cover percentage of the attack on the plant organs, "f" means the number of plant organs framed in a certain percentage and "b" represents the total number of affected plants.

## RESULTS AND DISCUSSIONS

By applying the serologic test DAS-ELISA (Table 1) we observed that only six of twenty cultivars were not infected with Plum pox virus: Tita (0,443), Tuleu Timpuriu (0,441), Diana (0,505), Flora (0,443), Vangerka jubilejnija (0,556) and Minerva (0,562). Values for the concentration of the viral protein in leaves smaller than 1,000 indicated the negative response of the tested plants to the viral infection.

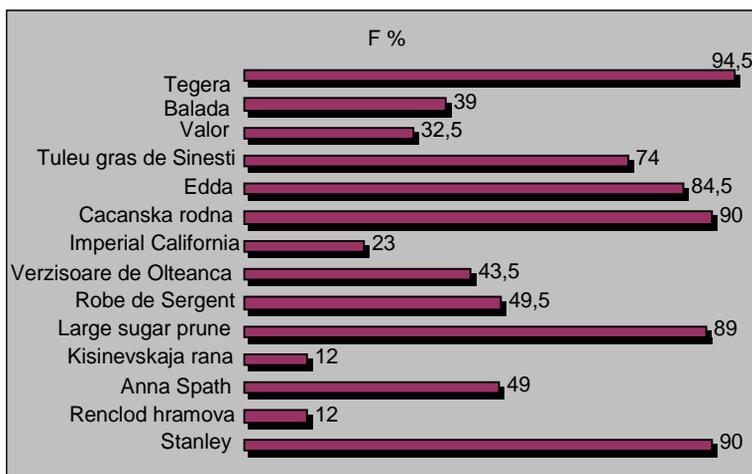
Table 1

The evidency of the Plum pox virus at plum cultivars through the serologic method

| No. Crt.          | The cultivar           | DAS-ELISA                   |                 |
|-------------------|------------------------|-----------------------------|-----------------|
|                   |                        | Values recorded at one hour | Sample reaction |
| 1                 | Tita                   | 0,443                       | -               |
| 2                 | Stanley                | >3,000                      | +               |
| 3                 | Renclod hramova        | 1,479                       | +               |
| 4                 | Tuleu timpuriu         | 0,441                       | -               |
| 5                 | Anna Spath             | 1,882                       | +               |
| 6                 | Kisinevskaja rana      | 3,258                       | +               |
| 7                 | Large sugar prune      | 3,258                       | +               |
| 8                 | Diana                  | 0,505                       | -               |
| 9                 | Robé de Sergent        | >3,000                      | +               |
| 10                | Verzişoare de Olteanca | 2,697                       | +               |
| 11                | Imperial California    | >3,000                      | +               |
| 12                | Čačanska rodna         | 2,125                       | +               |
| 13                | Flora                  | 0,443                       | -               |
| 14                | Edda                   | 2,220                       | +               |
| 15                | Tuleu gras de Sineşti  | 1,893                       | +               |
| 16                | Vangerka jubilejnija   | 0,556                       | -               |
| 17                | Minerva                | 0,562                       | -               |
| 18                | Valor                  | 1,126                       | +               |
| 19                | Balada                 | 3,257                       | +               |
| 20                | Tegera                 | >3,000                      | +               |
| Positive whitness |                        | >3,000                      | +               |
| Negative whitness |                        | 0,4550,464                  | -               |

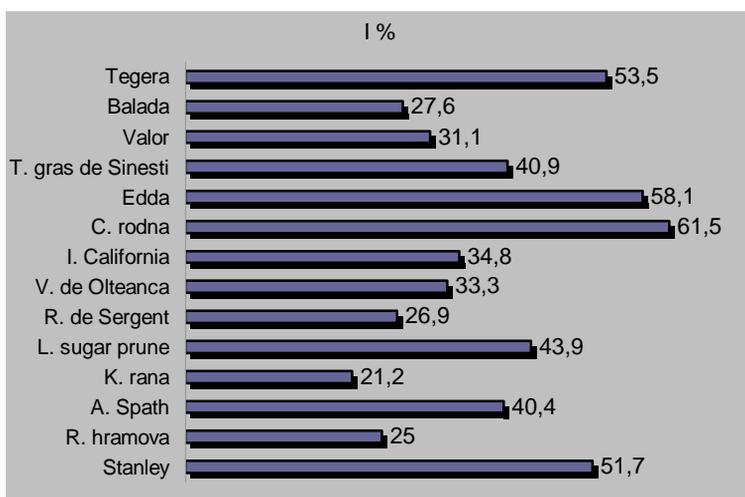
Note: - = healthy; + = infected

The frequency of the attack on leaves (figure 2) was very variable, the values oscillated between 12% and 94,5%. Low values were recorded at Renclod hramova, Kisinevskaja rana cultivars (12%) and very high fin case of four cultivars: Tegera (94,5%), Čačanska rodna, Stanley (90%) and Large sugar prune (89%) and Edda (84,5%).



**Fig. 2.** The frequency of the attack (%) of PPV on leaves

The limits between the values of the intensity attack oscillated were 21,2% (Kisinevskaja rana) and 61,5% (Čačanska rodna) (figure 3).



**Fig. 3.** The intensity of the attack (%) of PPV on leaves

According the interpretation scale for the values of the attack degree: AD % = 1-10 % week attack; AD % = 10-20 % medium attack; AD % = 20-30 % powerful attack; AD % > 30 % very powerful attack, we observed a week attack in case of three cultivars: Renclod hramova (3%), Kisinevskaja rana (2,5%), Imperial California (8%), a medium attack on the leaves of the cultivars Valor (10,1%), Balada (10,7%), Robé de Sergent (13,3%), Verzisoare de Olteanca (14,5%), Anna Spath (19,8%) and a very powerful attack in case of the cultivars

Tuleu gras de Sinești (30,2%), Large sugar prune (39,1%), Edda (49,1%), Tegera (50,6), Stanley (51,7%) and Čačanska rodna (55,4%) (figure 4).

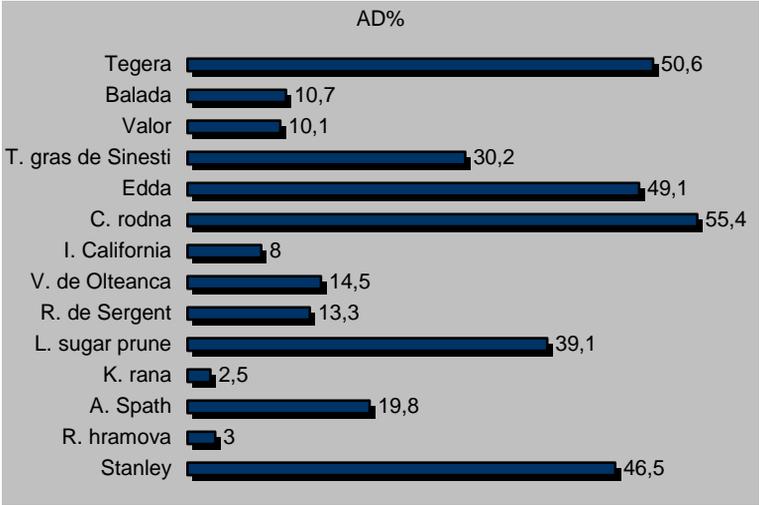


Fig. 4. The attack degree (%) of PPV on leaves

On fruits the attack degree of Plum pox virus was very small (values of the AD% were between 0,1% and 6,7%). Thus, of the five plum cultivars with characteristic symptoms on fruits, Edda was the most affected (6,7%) (figure 5).

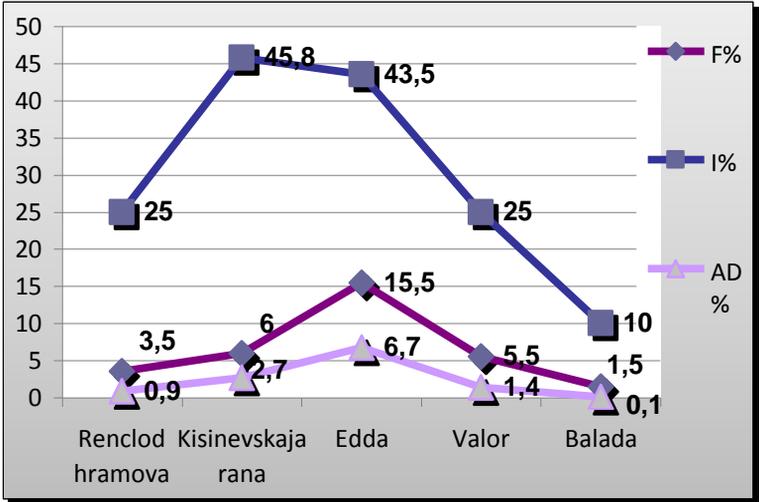


Fig. 5. The frequency, intensity and the attack degree of PPV on fruits

## CONCLUSIONS

1. The plum cultivars Tita, Tuleu timpuriu, Diana, Flora, Minerva and Vangerka jubilejnija proved not to be infected with Plum pox virus.

2. The attack degree of the virus on leaves was proved to be very powerful for six plum cultivars: Tuleu gras de Sinești, Large sugar prune, Edda, Stanley, Tegera and Čačanska rodna (the AD% values were higher than 30%).

3. The plum cultivars Renclod hramova, Kisinevskaja rana, Valor, Balada and Edda presented symptoms on fruits too, but the attack degree was less important (the AD% values were under 10%) by comparing him with the attack on leaves.

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