

**Faculty of Agriculture  
Goce Delcev University - Stip**



**2<sup>nd</sup> INTERNATIONAL MEETING  
AGRISCIENCE & PRACTICE  
(ASP 2019)**

**BOOK OF ABSTRACTS**

**12<sup>th</sup> April 2019  
Stip, Republic of North Macedonia**

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**FACULTY OF AGRICULTURE  
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12 April 2019, Stip, Republic of North Macedonia**

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# **PLANT PROTECTION**

**ONE STEP REAL-TIME POLYMERASE CHAIN REACTION USING FOR THE DETECTION OF PLUM POX POTYVIRUS** Cvetanka Kulukovska<sup>1\*</sup>, Emilija Arsov<sup>2</sup>, Sasa Mitrev<sup>2</sup>

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**Abstract**

Plant viruses are group of pathogens that cause important loses in different fruit production and they have great economic importance. They are obligate parasite forms and for their replication they used plant cells. One of the most important virus on the fruit is *Prunus* species, and the one that causes great economic losses is *Plum pox virus* (PPV), causal agent of Sharka disease. *Plum pox potyvirus* (PPV) is a filamentous virus that can be a part of phloem tissue in fruit-production species of *Prunus*, including apricots, peaches and plums. Since its discovery, Sharka has been considered as a calamity in plum orchards. In highly susceptible plum varieties presented in Macedonia, such as Požegača and Stenlej, PPV causes a premature fruit drop and reduces fruit quality, which leads to total yield loss. The same symptoms and loses are obviously in the peach and cherry garden. Eight PPV strains (PPV-M, PPV-D, PPV-EA, PPV-C, PPV-Rec, PPV-W, PPV-T and PPV-CR) have been recognized so far. Three major strains (PPV-M, PPV-D and PPV-Rec) are the most widely dispersed and occur frequently in many European countries. Other strains are of minor importance due to their limited host preferences or geographic distribution. In our research, plum hosts from several variety of plums were included in laboratory test analyses, such as plant parts (phloem, buds, flowers, leaves and fruits) and parts of them in different periods of the year (spring, summer and winter period 2017/18). The presence or absence of symptoms were considered for comparison. DAS-ELISA and Real Time PCR molecular techniques are included to confirm the presence and concentration of PPV in different plant material (leaves, stem, flower and fruit). By usage of DAS-ELISA tests, and a universal set of antibodies (BIOREBA), has proved the presence of virus of plum pox in all examined samples, especially from samples collected in spring, and in winter and early spring season, the virus status is on lower level. Testing found high concentrations of viral antigens in plant samples (OD 1.485 - 1.556, for 30 min). Results analysis from One step Real Time RT-PCR, show specific product for PPV that generate specific FAM-labeled amplification curve, and the Ct of the amplification curve is lower to 35 for positive samples. Total number of 10 DAS-ELISA positive samples (plum leaves), were confirmed with Real Time PCR with amplification curve with Ct≤35.

**Key words:** *Plum pox virus*; Sharka disease; DAS-ELISA, RT-PCR, PPV-M and PPV-D

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