

# Molecular and serological studies on an egyptian isolate of plum pox potyvirus

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## ABSTRACT

In this study, an Egyptian isolate of plum pox potyvirus (PPV), isolated from El-Amar apricot trees, was propagated on *Nicotiana benthamiana* for virus purification using a modified procedure was applied. Ultraviolet absorption spectrum of the purified virus showed a typical curve of nucleoprotein with an  $A_{260/280}$  ratio ranged from 1.258 to 1.280. A yield of the purified virus ranging from 20 to 22 mg/kg virus-infected leaf tissues was obtained. The electron microscopy of the purified virus preparation, negatively stained with 2% uranyl acetate, proved the presence of flexuous filament virus particles (700-725 x 13-15 nm). The molecular weight of PPV-coat protein (CP) was about 37 KDa when estimated by 12% SDS-PAGE. ssRNA of about 10 kb was extracted from the purified PPV preparation. Polyclonal antibodies against PPV were produced and the antiserum titer of three bleedings were determined by indirect-enzyme linked immunosorbent assay (I-ELISA). The IgGs specific to the PPV virions (intact virus particles) were purified and N- and C-terminal specific IgGs were selected and successfully used to distinguish the CP subunits of PPV-El-Amar isolate via western blot analysis. From PPV-infected leaf tissues, cDNA products were amplified via immunocapture-reverse transcription-polymerase chain reaction (IC/RT-PCR) using two specific primers for 3'-terminal region of PPV-cp gene. This was followed by restriction endonuclease analysis. The results revealed that the virus isolate under investigation is a strain of PPV and belongs to either M serotype due to the absence of *Rsa I* restriction site.

**Key words:** Plum pox potyvirus (PPV), purification, polyclonal antibodies, Western blotting, ELISA, Immunocapture/reverse transcription-polymerase chain reaction IC/RT-PCR, restriction endonuclease analysis.

## INTRODUCTION

Plum pox potyvirus (PPV), the causal agent of plum pox disease (sharka), is a member of family potyviridae, genus potyvirus (Brunt *et al.*, 1996). Sharka disease

is a series disease affecting stone fruit trees (SFTs) in Europe and Mediterranean regions. Apricot, peach and plum are the most important SFTs grown in Egypt, and are susceptible to infection with several viruses. Among such viruses, PPV is causing severe