

Serological and molecular characterization of an Egyptian isolate of banana-cucumber mosaic cucumovirus

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ABSTRACT

In this work, banana-cucumber mosaic cucumovirus (Banana-CMV) was isolated and purified from leaves of banana plant cv. Maghraby naturally exhibiting banana mosaic disease (BMD)-characteristic symptoms, collected from El-Kalubia governorate. The electron microscopy of the purified virus preparations showed spherical virus particles with a diameter of about 30 nm and a coat protein of about 26.5 kDa. The ultrathin sections of virus-infected banana leaves showed the presence of spherical virus-like particles in the cytoplasm of infected cells. In addition, flexuous virus like particles were also found associated with BMD. For the production of polyclonal antibodies (PABs) specific to Banana-CMV, the virus was purified from Banana-CMV-infected squash plants 3 weeks post virus inoculation using polyethylene glycol-8000. The antiserum was cross absorbed by the healthy banana proteins followed by purification of IgGs and labeling with alkaline phosphatase. An enzyme-linked immunosorbent assay (ELISA) kit was then developed and successfully used for Banana-CMV diagnosis. In addition, the coat protein gene of the isolated virus was detected via reverse transcription-polymerase chain reaction (RT-PCR) using two specific oligonucleotides. The amplified gene was confirmed by nested-PCR technique using two internal oligonucleotides as primers.

Key words: Banana, Banana-CMV, coat protein, ELISA, PCR, nested-PCR, polyclonal antibodies.

INTRODUCTION

Banana mosaic disease (BMD) caused by banana-mosaic cucumovirus (Banana-CMV) is one of the most important viral diseases affecting banana in Egypt (Allam *et al.*, 1995). BMD was first reported in the Philippines in 1926 (Stover, 1926). It has been called various names: infectious chlorosis, heart rot, mosaic disease and virus sheath rot. The severe form of the disease occurs sporadically wherever bananas are grown, but is not a major cause of loss

(Smith, 1972, Joshi and Joshi, 1976, Mali and Deshpande, 1976 and Mali and Rajegore, 1980).

The virus was isolated and identified as isometric particles with an average of about 30 nm in diameter by Hafner (1992) in Australia. CMV is transmitted in a non-persistent manner by aphids (Becquer and Bencomo, 1974; Retuerma, 1982; Patel and Mali, 1983; El-Afifi, 1984), and insecticidal sprays are of a limited value in controlling its spread (Walkey and Pink, 1984).