Purification, serology and molecular detection of Egyptian isolates of banana bunchy top babuvirus and faba bean necrotic yellows nanovirus

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ABSTRACT

Two isolates of faba bean necrotic yellows nanovirus (FBNYV) and banana bunchy top babuvirus (BBTV) were purified. Cellulase added to the extraction buffers (EB) enhanced extraction of both viruses. An acidic pH was used for FBNYV extraction. Several trials for purification of BBTV indicated that alkaline condition surpassed acidic condition in virus extraction. Polyethylene glycol and NaCl were used to concentrate both viruses. Electroelution coupled with freezing and thawing were employed in further purification steps. Purified FBNYV yielded 677 µg/ g tissue and with A 260/280 ratio of 1.43. Purified BBTV yielded 12.9 µg /g tissue and with A 260/280 ratio of 1.3. Both viruses had a single-capsid protein of 20 kDa.

An antiserum raised for the intact virions of FBNYV detected the purified virus and was able to cross react with BBTV in dot blotting immunobinding assay (DBIA) and in western blotting (WB). Antiserum for the whole virions of BBTV detected the virus in its vector Pentalonia nigronervosa as well as in infected banana tissues. This antiserum also cross-reacted with FBNYV in DBIA and in WB but failed to react with its own antigen of BBTV. On the otherhand, an antiserum raised for the coat protein of BBTV was able to detect both FBNYV and BBTV in WB. Such results suggest the presence of continuous epitopes in FBNYV virions and discontinuous epitopes in BBTV virions.

Polymerase chain reaction (PCR) experiments indicated that primers for the coat protein and replicase genes of BBTV amplified products in FBNYV genome similar to those produced in BBTV genome. Similar results were obtained upon using immunocapture (IC) PCR (IC-PCR) technique; indicating strong serologic relationships between the two viruses. The use of specific BBTV primers for nanovirus detection is of a great value in sensitivity and suitable for large scale testing.

Key words: *Purification, serology, molecular detection, BBTV, FBNYV.*

INTRODUCTION

BBTV is considered the most serious virus disease affecting banana worldwide. The disease was first recognized in Fiji in 1889, Taiwan in 1890, in Egypt 1901, and in Australia. 1913 (Fahmy, 1927; Wardlaw, 1961). It has spread and caused devastating problems in many countries including Fiji in 1927 and India and