Kalanchoë blossfeldiana, a New Host for *Sonchus Yellow Net Virus*

Inge Bouwen, Cor D. Schoen, Emmy van Balen, and René A.A. van der Vlugt
Plant Research International, Business Unit Bio-interactions and Plant Health
P.O. Box 16, NL-6700 AA Wageningen, The Netherlands

Abstract

The agent causing chlorotic spots in *Kalanchoë blossfeldiana* 'Isabella' was investigated. A virus isolated from this naturally infected kalanchoë was mechanically transmissible to several indicator plants. Observation of suspension preparations in the electron microscope revealed rhabdovirus-like particles. On the basis of symptoms on indicator plants, serology, electron microscopy, molecular characterisation and back inoculation to *K. blossfeldiana* 'Isabella', the causal agent was identified as an isolate of *Sonchus yellow net virus* (SYNV). This is the first report of an ornamental plant species naturally infected by SYNV.

INTRODUCTION

Sonchus yellow net virus (SYNV) is a nucleorhabdovirus known to infect one or more members of the plant families Chenopodiaceae, Compositae, and Solanaceae (Jackson & Christie, 1979). Lettuce is the only host of economic importance (Falk et al., 1986).

In recent years, plants of *Kalanchoë blossfeldiana* showing small chlorotic spots in the top leaves were regularly found. One of these plants, *K. blossfeldiana* 'Isabella', was tested negative for *Kalanchoë latent virus*, *Kalanchoë mosaic virus*, and *Tomato spotted wilt virus* in enzyme-linked immunosorbent assay (ELISA). Symptoms indicated that *Kalanchoë top spotting virus* could be involved. Using indicator plants, serology, electron microscopy, molecular characterisation and back inoculation to *K. blossfeldiana* 'Isabella', the agent causing the chlorotic spots in *K. blossfeldiana* 'Isabella' was identified.

RESULTS AND DISCUSSION

A plant of *Kalanchoë blossfeldiana* 'Isabella' showing small chlorotic spots in the top leaves was received from the Naktuinbouw, Roelofarendsveen, the Netherlands. In initial experiments the agent could be transmitted mechanically to *Chenopodium quinoa*, *Nicotiana glutinosa*, and *N. occidentalis* P1. In preparations of *N. occidentalis* P1 for observation in the electron microscope bacilliform particles were found. The particles had a clear striping and spikes on the outer membrane. These are characteristics for members of the *Rhabdoviridae*.

The rhabdovirus found in *K. blossfeldiana* 'Isabella' (KRV), and an isolate of SYNV, kindly supplied by A.O. Jackson, Department of Entomology, University of California, Berkeley, USA, were inoculated to a range of indicator plants. This range consisted of *C. quinoa*, *N. benthamiana*, *N. clevelandii*, *N. glutinosa*, *N. occidentalis* P1, *N. rustica*, and *N. tabacum* 'White Burley'. Similar symptoms were obtained for KRV and SYNV. There were local infections on *C. quinoa*, and local and systemic infections on all the tobacco species, except for *N. tabacum* 'White Burley', in which no infections were obtained.

KRV was purified from *N. benthamiana* and an antiserum was prepared. The isolate of KRV was compared with isolates of *Cereal chlorotic mottle virus*, *Eggplant mottled dwarf virus* (both kindly supplied by B.E.L. Lockhart, Department of Plant Pathology, University of Minnesota, St. Paul, USA), *Potato yellow dwarf virus* (kindly supplied by B.W. Falk, University of Florida, Agricultural Research and Education Center, Belle Glade, USA), and SYNV in DAS-ELISA using the KRV antiserum. A clear positive reaction was only obtained with samples infected with either KRV or SYNV. Thereupon, similar samples infected with KRV were tested in TAS-ELISA using an antiserum to SYNV, kindly supplied by the "Deutsche Sammlung von Mikroorganismen

und Zellkulturen GmbH" (DSMZ), Braunschweig, Germany. Also here clear positive results were obtained.

Suspension preparations of infected kalanchoë and indicator plants, and purified virus preparations were studied in the electron microscope. Bacilliform and bullet-shaped particles were found. The particle size was about 76 x 243 nm. In ultra-thin sections of *N. clevelandii* infected with KRV the rhabdovirus-like particles were mainly found in the perinuclear space. These results indicate that KRV is a nucleorhabdovirus.

For the molecular characterisation of KRV, PCR primers were designed based on the nucleocapsid encoding region of the sequence of SYNV (Acc. no. L32603). RT-PCR on RNA of KRV and SYNV resulted in DNA fragments of about 550 nucleotides. Sequencing of the PCR fragment of KRV revealed an overall nucleotide sequence homology of 81.9% and a deduced amino acid sequence homology of 97.2%.

To verify that the rhabdovirus initially transferred to *N. occidentalis* P1 is really the agent causing chlorotic spots in kalanchoë, it was inoculated back to virus-free *K. blossfeldiana* 'Isabella', kindly supplied by the Naktuinbouw. This back inoculation resulted in vein chlorosis and chlorotic spots in the youngest leaves. Similar symptoms were obtained when SYNV was inoculated to *K. blossfeldiana* 'Isabella'. All these infections of *K. blossfeldiana* 'Isabella' were confirmed by ELISA using the KRV antiserum.

The results obtained by using indicator plants, serology, electron microscopy, molecular characterisation and back inoculation to *K. blossfeldiana* 'Isabella', all confirm that the agent causing chlorotic spots in *K. blossfeldiana* 'Isabella' is an isolate of *Sonchus yellow net virus*. *Kalanchoë blossfeldiana*, a member of the Crassulaceae, is a new host for SYNV. It is the second host of economic importance, and the first ornamental plant species to be found naturally infected by SYNV.

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Literature Cited

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