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Short Communication

## FIRST REPORT OF *TOMATO BLACK RING VIRUS* (TBRV) IN THE NATURAL INFECTION OF *SAMBUCUS NIGRA* IN POLAND

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Black elderberry (Sambucus nigra) is wild grown shrub in Poland. S. nigra occurs in the whole country on various wast lands, in parks, forests, old ruins but also on fields and around home gardens. In the late spring, virus like symptoms very often were observed on this shrubs. Symptoms consisted of chlorotic flecks, vein clearing, some oak-leaf pattern on leaves. Studied virus was isolated in the middle of May 2002 near Warsaw city, from black elderberry plants with symptoms of strong inhibition of leaves development. The host range and symptoms for the studied virus were determinated by mechanical inoculation of various plant species with the sap of infected Chenopodium quinoa. Reactions of indicator plants to the virus are listed in the table 1. Mechanical inoculation of Chenopodium ssp. resulted in chlorotic or necrotic lesions, with subsequent systemic deformations and necrosis of top. Nicotiana tabacum cvs. Samsun and Xanthi nc reacted with characteristic local and systemic ring spot or pattern lines but later they recovered. These symptoms were similar to those caused by Tomato black ring virus (TBRV) from cucumber (Pospieszny et al. 2003). Seven days after inoculation, C. quinoa plants were checked for the presence of virus by electron microscopy (EM) observations. In the sap spherical virions, so called empty and full (Fig. 1), and sometimes characteristic tubules containing isometric particles were observed.

The virus particles purified from *C. quinoa* were sedimented in a sucrose density gradient as three zones opalescent in a transmitted light. Electron microscopy observation showed that top zone contained empty capsids.

Virus was also identified serologically, by ELISA test, using antisera against TBRV and *Beet ringspot virus* (BRSV). Positive reaction were observed only with TBRV antiserum.



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Table 1. Host range and symptoms for TBRV from Sambucus nigra

Plant species	Symptoms
Chenopodium quinoa	Lch-ns; Sd, Sn
C. amaranticolor	Lch-ns; Sm, Sd
C. album	Lchs; Sm, Sd
C. murale	Lchs; Sn
C. ficifolium	(Lchs); Sm
Nicotiana tabacum cv. Xanthi nc	Lrs; Srs, R
N. tabacum cv. Samsun	Lrs; Srs, R
N. clevelandii	Lrs; Sns, R
N. glutinosa	(Lchs); Sm
N. debneyi	Lrs; Schs, R
N. affinis	(Lchs); Srs, R
N. benthamiana	(Lchs); Sm
Petunia hybrida	–; Sm, s
Datura stramonium	-; Sm
Nicandra physaloides	-; Sm
Lycopersicon esculentum	–; Schs, Sm, Sd, (s)
Phaseolus vulgaris	Lchs; Sm
Pisum sativum	(Lchs); Sm, Sn
Lupinus albus	-
Synapsis alba	-; s
Brassica rapa	-
Spinacia oleracea	(Lchs); Sm
Tetragonia expansa	(Lchs); Sm, Sd
Cucumis sativus	–; Sm, (s)
Lactuca sativa	-; s
Zinnia elegans	-; s
Ammi maius	-; s

Lchs=local chlorotic spots, Lch-ns=local chlorotic-necrotic spots, Lns=local necrotic spots, Lrs=local ring spots, Sm=systemic mosaic, Schs=systemic chlorotic spots, Srs=systemic ring spots or patern lines, Sd=systemic deformation; Sns=systemic necrotic spots, Sn=necrosis of top, R=recovery, s=symptomless infection, ()=symptoms appeared sporadically, – = no symptoms or no virus

This identification was confirmed by immunocapture-reverse transcription polymerase chain reaction (IC-RT-PCR) with primers designed by Le Gall et al. (1995). The antibodies against TBRV-ED were used for trapping of virus particles from plant sap. The results of IC-RT-PCR for both, TBRV from *S. nigra* and TBRV from cucumber (Pospieszny et al. 2003) were 300 nucleotides (Fig. 2).

From purified virus preparation RNA was isolated and separated by electrophoresis in 1% agarose gel (Pospieszny et al. 2003). Genomic RNAs of TBRV from *S. nigra* migrated as two bands, typically for nepoviruses (Fig. 3).

Several authors in different parts of the world found various viruses in *S. nigra* (Schmelzer 1966; Stefanac 1969; Hansen and Stace-Smith 1971; Horvath et al. 1974; Polak et al. 1990; Cooper 1993). It was shown that elderberries could carry viruses which may cause serious losses in stone fruit cultivars (Gilmer and Kelts 1968; Lister 1964; Schmelzer 1966). In Poland, TBRV was found in the number of species of cultivated and wild plants (Pospieszny et al. 2003) but it is the first report concerning this virus in *Sambucus nigra*. Black elderberry is a commonly occurring

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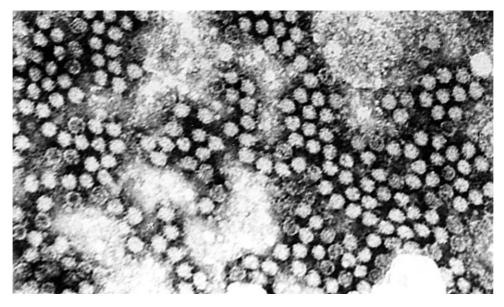


Fig. 1. Virions of Tomato black ring virus from Sambucus nigra

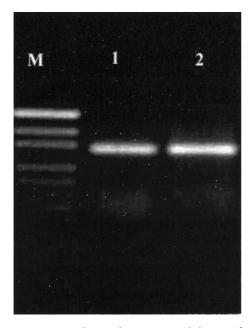


Fig. 2. Electrophoretic mobility of IC-RT-PCR product Lane M:DNA leader pVC 19 DNA/Mspl (501, 404, 331, 242, 190, 147, 111, 67, 34) Lane 1: TBRV – S. nigra Lane 2: TBRV - potato

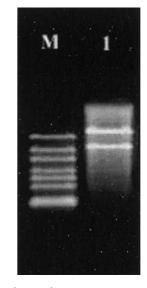


Fig. 3. Electrophoretic separation of viral RNA on 1% agarose gel Lane M: RNA Leader (6 kb, 4 kb, 3 kb, 2 kb, 1.5 kb, 1 kb, 0.5 kb, 0.2 kb) Lane 1: TBRV-S. nigra

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shrub in Poland but its role as a natural source of TBRV seems to be limited due to its relative low efficiency of transmission by nematode.

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