

Short Note

The First Record of Nucleopolyhedrovirus Isolated from the Satin Moth *Leucoma salicis* L. (Lepidoptera, Lymantriidae) in Turkey

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A nucleopolyhedrovirus was isolated and characterized for the first time from *Leucoma salicis* L. (Lepidoptera, Lymantriidae) in Turkey. The virus was observed in populations of *L. salicis* in Güntüşhane. The dimensions of the polyhedrae fell between 2.08 ± 0.31 (1.51-2.64) μm (N=50). Virions contain 2 to 15 nucleocapsids per virion as seen in cross-section of polyhedrae. The sizes of the viral particles ranged between 250-290 x 32-40 nm. The virus was determined as a Turkish isolate of *Leucoma salicis* nucleopolyhedrovirus (LesaNVPV-TR).

Key words: *Leucoma salicis* L. (Lepidoptera, Lymantriidae), nucleopolyhedrovirus, Turkish isolate, biological control.

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The satin moth *Leucoma salicis* L. (Lepidoptera, Lymantriidae), previously known as *Stilpnotia salicis*, is a serious defoliator occurring throughout Europe and Asia (LIPA & ZIEMNICKA 1996). It is also an important pest in Turkey (ÇANAKÇIOĞLU 1983). Satin moth larvae feed on all species of poplar and willow (*Populus* spp.), but also on oak and crabapple (JAKUBOWSKA *et al.* 2005). Severe feeding damage results in reduced growth of stems and finally tree mortality (LANGOR 1995). Increasing problems with resistance of this pest to most commonly used synthetic insecticides has spurred the search for alternative pest management strategies that reduce reliance on synthetic insecticides. Fortunately, *L. salicis* populations are reduced by a natural nucleopolyhedrovirus (NPV) (Baculovirus) disease (ZIEMNICKA 1981). This virus is capable of causing disease in the larvae of *L. salicis*, but does not harm other animals or plants. Nucleopolyhedroviruses represent an effective, selective and safe biological control agent (GREATHEAD 1976). The best results with nucleopolyhedroviruses have been achieved against lepidopteran pests, for example *L. salicis* using LesaNVPV (ZIEMNICKA 1981; 2000). Several important studies have been carried out on the Polish isolate of *Leucoma salicis* nucleopolyhedrovirus (STROKOVSKAYA *et al.* 1996;

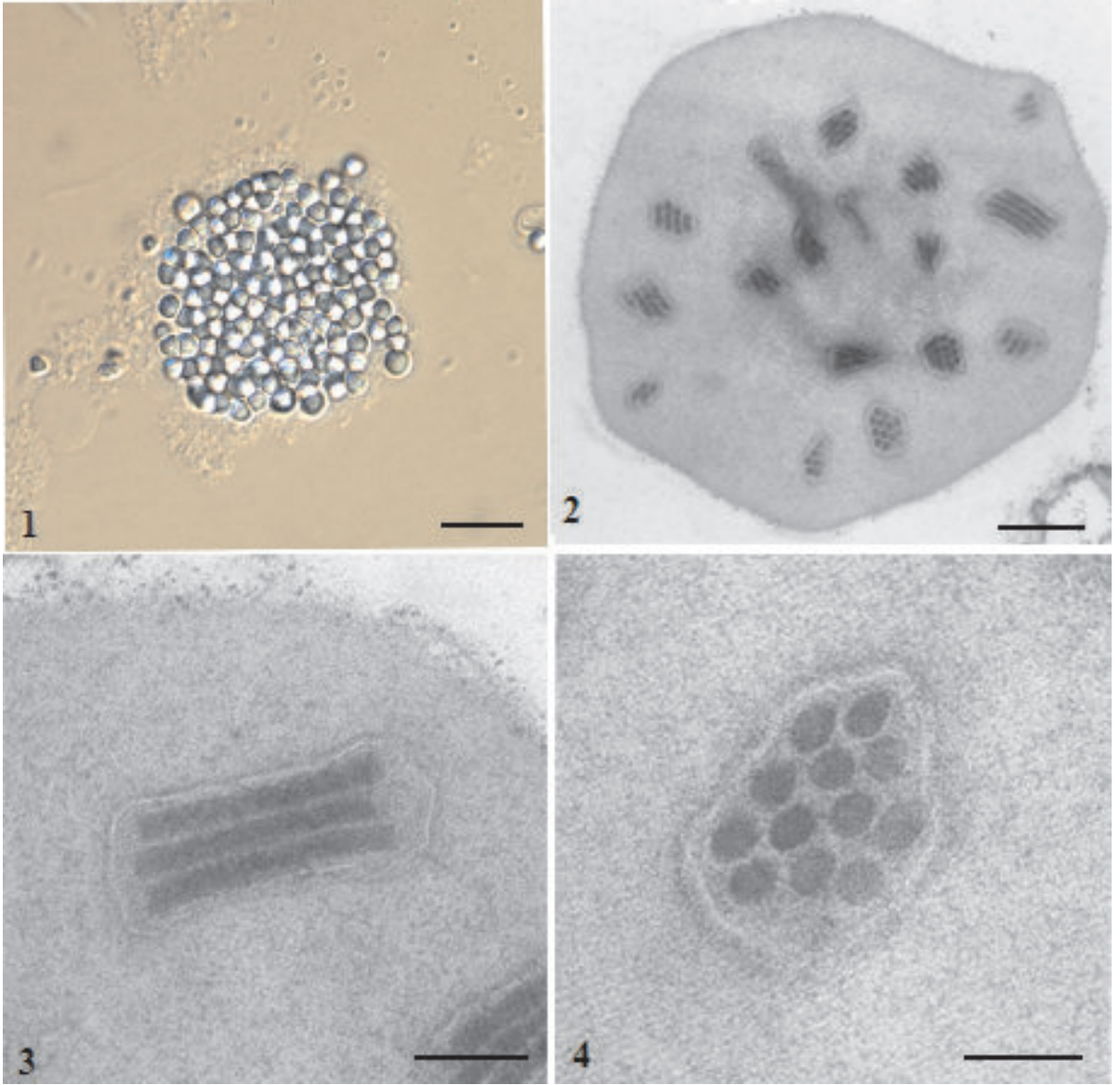
LIPA & ZIEMNICKA 1996; JAKUBOWSKA *et al.* 2005). Some isolates of this virus were recorded in part of Europe (WEISER *et al.* 1954; SKATULLA 1985) but there is no record of the isolation and characterization of this virus from Asia. Asia is a potential source of new and interesting NPV strains (MURILLO *et al.* 2001). Some strains of nucleopolyhedrovirus isolated from different geographic localities may present better insecticidal activities, which make them more suitable for host control and show important differences in biological activity (MURILLO *et al.* 2001).

In the present study, the isolation and characterization of a novel isolate of *Leucoma salicis* nucleopolyhedrovirus (LesaNVPV-TR) from Turkey are presented for the first time.

Material and Methods

Collecting infected larvae of *L. salicis*

Virus-infected larvae of *L. salicis* were observed on wild poplar foliage in Güntüşhane located in South Central Province of the Black sea Region of Turkey. The infected larvae were transferred to col-



Figs 1-4. Fig. 1 – Polyhedra of *Neodiprion sertifer* nucleopolyhedrovirus, light microscope, bar = 10 μ m; Fig. 2 – Section of a polyhedra with virions containing multiple rod-shaped nucleocapsids, bar = 300 nm; Fig. 3 – Longitudinal section of virion, bar = 125 nm; Fig. 4 – Cross section of virion, TEM, bar = 200 nm.

lection tubes using sterile forceps. A fresh pair of forceps for each new sample was used to prevent cross-contamination of material. Collected samples were brought to the laboratory as soon as possible and stored at -20°C (HUNTER-FUJITA *et al.* 1998).

Determination of viral infection

The infected larvae were dissected, and polyhedra were observed under a light microscope. In addition, the presence of viral infection was confirmed with Giemsa staining (HUNTER-FUJITA *et al.* 1998). Polyhedrae from the LesaNPV-TR isolate were examined by transmission (TEM) electron microscopy. Different portions of infected larvae were fixed in 2.5% glutaraldehyde in 0.1 M cacodylate buffer (pH 7.4) for 1-2 h, rinsed in

cacodylate buffer, postfixed in reduced OsO_4 according to KARNOVSKY (1971) (a fresh 1:1 mixture of 2% OsO_4 and 3% $\text{K}_4[\text{Fe}(\text{CN})_6]$) for 1.5 h, rinsed in cacodylate buffer and dehydrated in an ethyl alcohol series prior to embedding in Spurr's resin (SPURR 1969). Thin sections were mounted on Pioloform-coated copper grids which were stained with saturated uranyl acetate (WATSON 1958) and Reynold's lead citrate (REYNOLD 1963). They were examined in a Philips 208 electron microscope.

Results and Discussion

The virus was detected in populations of *Leucoma salicis* in Gümüşhane. The infected larvae

were less active and dark-brown color. Their cuticula was easily broken. The majority of the dead larvae hang on poplar branches and leaves, characteristically attached with abdominal prolegs. Under the light microscope, a high number of polyhedral inclusion bodies (PIBs) formed by the virus was observed (Fig. 1). The dimension of the polyhedral inclusion bodies is $2.08 \pm 0.31 \mu\text{m}$ ($n=50$). PIBs vary in size from 1.51 to 2.64 μm and were usually polygonal in shape (Fig. 2).

In electron microscopy studies, virions contained 2 to 15 nucleocapsids per virion (Figs 3 & 4). The size of the viral particles was 270.71 ± 12.72 ($250\text{-}290$) \times 33.11 ± 3.48 ($32\text{-}40$) nm. In a recent detailed study on the Polish isolate of LesaNPV, ZIEMNICKA (1981) reported virions 306 ± 11 nm in length and 42.9 ± 2.1 nm wide. Similarly, WEISER *et al.* (1954) recorded virions $270\text{-}400 \times 100\text{-}150$ nm in size. Clear differences are seen among Turkish, Polish and Czech isolates. As ZIEMNICKA (1981) suggests, it is rather improbable that these differences are caused by the use of various techniques and perhaps two different virus strains occur in Turkey and Poland. This idea is supported by MURILLO *et al.* (2001). These authors suggest that some strains of nucleopolyhedrovirus isolated from different geographic localities may possess better insecticidal activities.

LesaNPV gives promising results in the above mentioned higher insecticidal activities (HUBER 1998). Several important studies have been carried out on *Leucoma salicis* nucleopolyhedrovirus isolates from Western Europe (LAMERIS *et al.* 1985; HUBER 1998) and Eastern Europe (STROKOVSKAYA *et al.* 1996; LIPA & ZIEMNICKA 1996; ZIEMNICKA 1981, 2000; JAKUBOWSKA *et al.* 2005). Some isolates were also recorded from different parts of Europe (WEISER *et al.* 1954; SKATULLA 1985). Unfortunately there is no record of natural isolation and characterization of this virus from Turkey as a different geographical locality from Asia, whereas Asia is a potential source of new and interesting NPV strains (MURILLO *et al.* 2001). It is known that some strains of nucleopolyhedrovirus isolated from different geographic localities may present better insecticidal activities, which make them more suitable for host control and show important differences in biological activity (MURILLO *et al.* 2001). For example, a Polish NPV isolate was about seven times more infectious than one from Yugoslavia (ZIEMNICKA 1981; LAMERIS *et al.* 1985; LIPA 1998).

In the present study, the isolation and characterization of a novel isolate of *Leucoma salicis* nucleopolyhedrovirus (LesaNPV-TR) from Turkey is presented for the first time. The results of this study will encourage scientists to compare the infectivity and systematics of LesaNPV isolates

from different parts of Europe and Asia at the molecular level, especially Polish and Turkish isolates, in order to find the most effective strain and understand their evolution.

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