

Isolation of *Olive latent virus 1* from Tulip in Toyama Prefecture[†]

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ABSTRACT

A virus whose coat protein gene had a high sequence homology with the coat protein gene of *Olive latent virus 1* was isolated from diseased tulip in Toyama Prefecture.

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Key words : *Olive latent virus 1*, coat protein gene, tulip.

Tulip necrosis disease is one of the most important diseases for tulip bulb production, and several reports describe the causal agent as *Tobacco necrosis virus* (TNV). Strains of TNV have been grouped into two distinct but related serotypes, A and D¹⁾. In Toyama Prefecture, viruses were isolated from diseased tulip and identified as isolates of TNV based on their biological and immunological properties⁵⁾. One isolate, Pare-P, isolated from tulip without typical necrotic symptoms in 1987, was further investigated to determine whether it belonged to the A- or D-strain of TNV.

A primer set was designed to amplify the coat protein (CP) region of both strains, 5'-dAAGACTCAACACATT-TCGATCG-3' as a forward primer and 5'-dAGCCTGTT-TCCCAGGATCCG-3' as a reverse primer. The amplified fragment (ca. 950 bp) was inserted into the pT7 Blue (Novagen) vector and sequenced.

The CP region of Pare-P consisted of 810 nucleotides encoding 270 amino acids. The nucleotide sequence had only 54-56% homology with TNV sequences registered in DDBJ/EMBL/GenBank, but had a remarkably high homology (93%) with *Olive latent virus 1* (OLV-1). Multiple alignment and a phylogenetic tree based on their deduced amino acid sequences showed the close relationship between Pare-P and OLV-1 (Fig. 1).

OLV-1 is a member of the genus *Necrovirus*³⁾ and has been isolated from symptomless olive²⁾ and chlorotic dwarf diseased citrus⁴⁾. The host range of these two isolates was examined in experimental herbaceous plants, but not in other plants such as tulip. The result shown in

Fig. 1 indicated that Pare-P was more closely related to OLV-1 rather than TNV. Thus, the Pare-P isolate was tentatively named the tulip isolate of OLV-1.

TNV-A and TNV-D antisera reacted very weakly with OLV-1⁴⁾, and the antiserum against the Pare-P isolate did

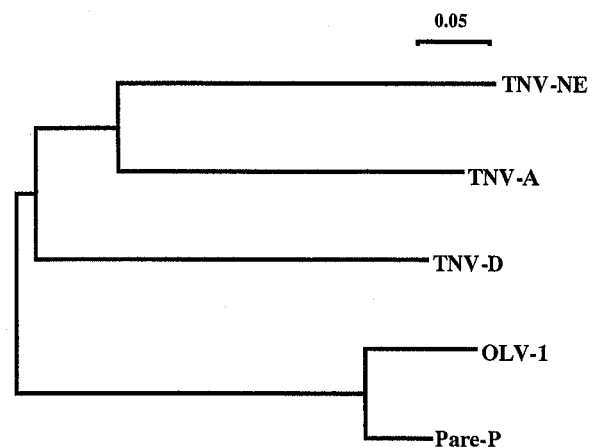


Fig. 1 Phylogenetic tree displayed by the NJplot after alignment by Clustal X. The bar indicates a distance of 0.05. Virus abbreviations and DDBJ/EMBL/GenBank data accession numbers are as follows : TNV-A=*Tobacco necrosis virus* A strain (M33002), TNV-D=*Tobacco necrosis virus* D strain (D00942), TNV-NE=*Tobacco necrosis virus* Nebraskan isolate (L04261), and OLV-1=*Olive latent virus 1* (X85989).

[†] The nucleotide sequence determined in this work appears in the DDBJ/EMBL/GenBank nucleotide sequence databases with the accession number AB061815.

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not react with TNV-A (unpublished data). This data agrees with the phylogenetic relationship among TNV strains and OLV-1 isolates shown in Fig. 1.

Pare-P was originally isolated from tulip with mottle or yellow streak symptoms⁵. Next, symptoms should be reproduced in tulip after inoculation with OLV-1 to confirm pathogenicity and to compare the biological properties of OLV-1 isolates.

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