



# Complete Genome Sequence of a *Lily virus X* Isolate from Japan

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**ABSTRACT** The complete genome sequence of *Lily virus X* (LVX), which infects lilies, was determined for the first time from lilies in Japan. As with previous reports, the genome of the Japanese LVX isolate lacked an AUG start codon for the triple gene block protein 3-like region.

*Lily virus X* (LVX) is a member of the genus *Potexvirus* in the family *Alphaflexiviridae* (1, 2). LVX possesses flexuous filamentous particles and a single-stranded positive-sense RNA genome. LVX was first reported in a symptomless lily (*Lilium formosanum*) from England (3) and is known to occur in the Netherlands and the United States (4, 5). To date, the complete genome of one LVX isolate from the Netherlands has been reported (6). Although the reported genome organization of LVX was similar to that of other members of the genus *Potexvirus*, it lacked an AUG start codon for the triple gene block protein 3 (TGBp3), one of the movement proteins (4, 6). In Japan, although LVX is known to be the causative agent of lily necrosis when associated with lily mottle virus (7) and has also been detected from symptomless lily cultivars (8), sequence information on Japanese LVX isolates has not been reported. Here, we report for the first time a complete genome sequence of LVX isolated from a lily in Japan.

Lily plants (*Lilium cv. Acapulco*) were collected in the Chubu District of Japan in 2016. Total RNA was extracted from leaves using Isogen reagent (Nippon Gene, Japan). A paired-end sequencing cDNA library was constructed from the extracted RNA using a TruSeq RNA sample prep kit v.2 (Illumina, USA) and sequenced using a MiSeq instrument (Illumina) and a MiSeq reagent kit v.2 (500 cycles). After quality control and trimming using Trimmomatic v.0.36 software (9), the reads were *de novo* assembled using Trinity v.2.3.2 software (10). The assembled contigs were subjected to a BLASTn search (11) against the GenBank database, and a contig showing sequence identity with LVX (GenBank accession number AJ633822) was obtained. To determine the complete genome sequence, the 5' and 3' ends of the genome were amplified by rapid amplification of cDNA ends (RACE) using a Gene Racer kit (Invitrogen, USA). The amplified fragments were cloned into pCR-Blunt II-TOPO vector (Invitrogen) and sequenced.

The complete genome sequence of the Japanese isolate of LVX (LVX-J) was 5,824 nucleotides (nt) long, excluding the poly(A) tail at its 3' end. It contained four open reading frames (ORFs) encoding RNA-dependent RNA polymerase (RdRp) (nt 73 to 3969), TGBp1 (nt 3999 to 4649), TGBp2 (nt 4649 to 4975), and coat protein (CP) (nt 5110 to 5715). The TGBp3-like region (nt 4893 to 5129) lacked an AUG start codon, as in the case of the previously reported isolate (6). The respective percent identities of RdRp, TGBp1, TGBp2, TGBp3, and CP with those of the reported LVX isolate (GenBank accession number AJ633822) were 99.4%, 98.8%, 99.7%, 99.6%, and 99.3% at the nucleotide level and 99.3%, 98.6%, 99.1%, 97.4%, and 99.0% at the amino acid level,

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respectively. These results indicate that LVX-J is closely related to the reported LVX isolate from the Netherlands. Because lilies are propagated vegetatively from bulbs, a genetically identical strain of LVX might be spread worldwide by LVX-infected lily bulbs.

**Accession number(s).** The complete genome sequence of *Lily virus X* isolate J has been deposited in the DNA Data Bank of Japan and GenBank under the GenBank accession number [LC335818](https://www.ncbi.nlm.nih.gov/nuccore/LC335818).

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## REFERENCES

1. Adams MJ, Antoniw JF, Bar-Joseph M, Brunt AA, Candresse T, Foster GD, Martelli GP, Milne RG, Zavriev SK, Fauquet CM. 2004. The new plant virus family *Flexiviridae* and assessment of molecular criteria for species demarcation. *Arch Virol* 149:1045–1060. <https://doi.org/10.1007/s00705-004-0304-0>.
2. Adams MJ, Candresse T, Hammond J, Kreuze JF, Martelli GP, Namba S, Pearson MN, Ryu KH, Vaira AM. 2012. Family *Alphaflexiviridae*, p 904–919. In King AMQ, Adams MJ, Carstens EB, Lefkowitz EJ (ed), *Virus taxonomy: ninth report of the International Committee on Taxonomy of Viruses*. Elsevier Academic Press, San Diego, CA.
3. Stone OM. 1980. Two new potexviruses from monocotyledons. *Acta Hort* 110:59–64. <https://doi.org/10.17660/ActaHortic.1980.110.5>.
4. Memelink J, van der Vlugt CI, Linthorst HJ, Derks AF, Asjes CJ, Bol JF. 1990. Homologies between the genomes of a carlavirus (lily symptomless virus) and a potexvirus (lily virus X) from lily plants. *J Gen Virol* 71:917–924. <https://doi.org/10.1099/0022-1317-71-4-917>.
5. Jordan RL, Guaragna MA, Van Buren T, Putnam ML. 2008. First report of a new potyvirus, *Tricyrtis virus Y*, and *Lily virus X*, a potexvirus, in *Tricyrtis formosana* in the United States. *Plant Dis* 92:648. <https://doi.org/10.1094/PDIS-92-4-0648A>.
6. Chen J, Shi YH, Adams MJ, Chen JP. 2005. The complete sequence of the genomic RNA of an isolate of *Lily virus X* (genus *Potexvirus*). *Arch Virol* 150:825–832. <https://doi.org/10.1007/s00705-004-0441-5>.
7. Hagita T, Sasaki J, Mukohara M. 2000. *Lily virus X* isolated from necrosis of the edible lily, *Lilium leichtlinii* var. *maximowiczii* Baker. *Ann Rept Plant Prot N Jpn* 51:98–103. (In Japanese with English summary.)
8. Kimura S, Goto M, Saito N, Fujiwara Y. 1990. Detection of *Lily virus X* (LVX) from several lily cultivars grown in Japan. *Res Bull Plant Protect Serv Jpn* 26:79–81. (In Japanese with English summary.)
9. Bolger AM, Lohse M, Usadel B. 2014. Trimmomatic: a flexible trimmer for Illumina sequence data. *Bioinformatics* 30:2114–2120. <https://doi.org/10.1093/bioinformatics/btu170>.
10. Haas BJ, Papanicolaou A, Yassour M, Grabherr M, Blood PD, Bowden J, Couger MB, Eccles D, Li B, Lieber M, MacManes MD, Ott M, Orvis J, Pochet N, Strozzi F, Weeks N, Westerman R, William T, Dewey CN, Henschel R, LeDuc RD, Friedman N, Regev A. 2013. *De novo* transcript sequence reconstruction from RNA-seq using the Trinity platform for reference generation and analysis. *Nat Protoc* 8:1494–1512. <https://doi.org/10.1038/nprot.2013.084>.
11. Altschul SF, Gish W, Miller W, Myers EW, Lipman DJ. 1990. Basic local alignment search tool. *J Mol Biol* 215:403–410. [https://doi.org/10.1016/S0022-2836\(05\)80360-2](https://doi.org/10.1016/S0022-2836(05)80360-2).