

Potyvirus Leek Yellow Stripe Virus

Scientific Name:

Potyvirus Leek Yellow Stripe Virus (Bos, 1978)

Synonyms:

Garlic mosaic virus, garlic yellow stripe virus, garlic yellow streak virus

Common Name(s)

Leek Yellow Stripe Virus (LYSV)

Type of Pest

Plant pathogenic virus

Taxonomic Position

Class: Not assigned, **Order:** Not assigned, **Family:** Potyviridae

Reason for Inclusion in Manual

Additional Pest of Concern

Pest Description

Allium species (garlic, onion, and leek) are grown commercially all over the world for consumption or ornamental decoration. *Leek yellow stripe virus* (LYSV) is the causal agent of yellow stripe disease in leek and mosaic disease in garlic. Severe epidemics of yellow stripe disease on leek in Germany in the 1950s, and twenty years later in the Netherlands catalyzed more intensive research into the viral causal agent of the disease (Bos, 1983). These investigations proved difficult, as the causal agents of yellow striping occurred in virus complexes that were challenging to characterize. The first official report of LYSV in commercial garlic was in 1987, but later investigations demonstrated the presence of the virus in New Zealand as early as 1981 (Lot et al., 1998)

The virus is a positive-sense single stranded RNA virus that houses its genetic material in a non-enveloped capsid (Davis, 2008a). The virus particles (Fig. 1) are flexuous (rod-like and bendable) particles 815 to 820nm long that aggregate end-to-end. The protein coat of the virus is 34 kDa with a buoyant density in a cesium chloride gradient of 1.326 g/cm³ (Bos, 1981).

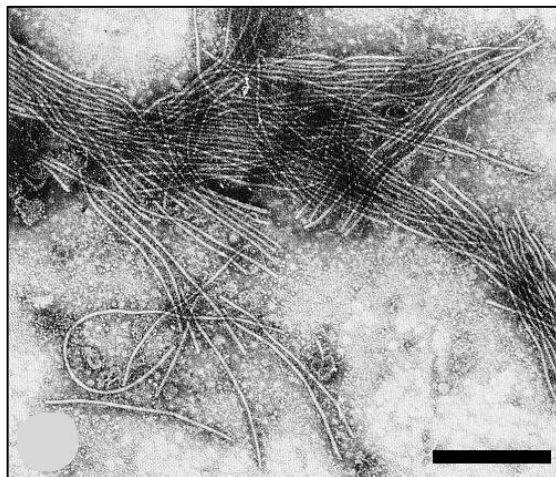


Figure 1. Electron micrograph of LYSV viral particles from infected leek. Photo courtesy of the Association of Applied Biologists.

Biology and Ecology

Leek Yellow Stripe Virus is a potyvirus specific to *Allium* species (Bos et al., 1978). The *Allium* potyviruses rarely cross over to infect new hosts, which eases the threat of contamination and spread to neighboring non-*Allium* hosts. LYSV is commonly found in nature in viral complexes with other *Allium* potyviruses and carlaviruses, such as onion dwarf yellow virus and garlic common latent virus. The appearance of LYSV in these viral complexes made initial identification and classification difficult (Barg et al., 1997).

Symptom expression is rare and haphazard in the field during summer months, but becomes increasingly apparent in the fall months. Autumn and winter crops can be completely infected (Bos et al., 1978). In the late 1970s, there was a shift from lighter-colored cultivars to darker-colored, “less sensitive” cultivars in commercial production. This led to a decrease in the incidence of LYSV in autumn crops. This apparent resistance has been credited to the thicker cuticular wax layer on the darker leek cultivars (Vandijk, 1993).

Symptoms and Signs

Leek: In leek, LYSV causes irregular yellow striping on the leaves (Fig. 2), particularly near the base, but not confined to this area. In certain instances, entire leaves can turn yellow (Bos et al., 1978). Diseased plants are drier with soft, limp, and/or deformed leaves (Barg et al., 1997). Symptoms observed in the field could be replicated in the greenhouse, and usually appeared within 14 days of inoculation (Lot et al., 1998).

Garlic: In garlic, LYSV causes light yellow striping on the distal part the leaves, which can lead to dwarfing of the entire plant (Fig. 3). The virus also causes bulbs to be smaller and malformed, which results in yield loss.

Allium crops infected with LYSV are more susceptible to weather conditions like frost, and do not keep well post-harvest (Bos et al., 1978).



Figure 2. Infected garlic leaves with chlorotic striping. Photo courtesy of Dr. Hanu Pappu.



Figure 3. Severely dwarfed garlic with necrosis on the leaf tips. Photo courtesy of Dr. Mike Pearson.

Pest Importance

Potyvirus rarely infect in isolation, and complexes of different strains and species are common. Infections of LYSV in the field can be devastating, sometimes approaching 100% on leek plots (Bos, 1983). Yield loss in garlic bulbs can approach 60% and climb to 84% when doubly infected with another potyvirus. Germination rate in garlic is also affected and can decline to 60% of normal when doubly infected with another potyvirus (Lot et al., 1998). Garlic exports in the United States were valued at approximately \$8.8 million in 2008, and world-wide production was valued at \$187 million. Significant losses are a possibility if the disease becomes widely established in the United States (NPAG, 2009).

Known Hosts

Major hosts:

Allium ampeloprasum var. *holmense* (great-headed garlic), *A. ampeloprasum* var. *porrum* (leek), *A. aperloprasum* var. *sectivum* (pearl onion), *A. longicupis* (wild garlic), and *A. stivium* (garlic) (Bos, 1981; Vandijk, 1993).

Minor hosts:

Allium cepa var. *cepa* (onion) and *A. cepa* var. *ascalonicum* (shallot) (Bos, 1981; Vandijk, 1993).

Experimental/Indicator Hosts:

Chenopodium amaranticolor (tree spinach), *C. quinoa* (quinoa), and *Celosia argentea* (silver cock's comb) (Bos, 1981; Vandijk, 1993).

Known vectors (or associated organisms)

Common aphid vectors of LYSV are the green peach aphid (*Myzus persicae*) and the black bean aphid (*Aphis fabae*) (Fig. 4, 5) (Lunello et al., 2002). Both species are found on every continent except Antarctica, and in over half of the 50 United States (CIE, 1963). These aphids are mono- or polyphagous depending on the growth stage. The youngest stages have a single primary host during the winter and spread to other hosts in the summer months as the nymphs grow. They both spread numerous plant viruses in addition to LYSV in a non-persistent manner during feeding (Blackman and Eastop, 2000).



Figure 4. (Left to right) Young nymph, adult wingless, and winged green peach aphids. Photo courtesy of Rothamsted Research.

Other aphids used to experimentally inoculate host plants include: *Rhopalosiphum maidis* (corn aphid, corn leaf aphid), *R. padi* (bird cherry oat aphid), *Schizaphis graminum* (greenbug), *Aphis gossypii* (melon aphid,

cotton aphid), *A. nerii* (oleander aphid), *Uroleucon sonchi* (sowthistle aphid), and *Hyperomyzus carduellinus* (Asian sowthistle aphid) (Lunello et al., 2002).

LYSV is also commonly associated with other *Allium* viruses, *i.e.*, it has a synergistic relationship with onion dwarf virus. Multiple investigations have demonstrated that plants express aggravated symptoms when infected with LYSV and one or more other viruses (Vandijk, 1993; Lot et al., 1998). Reverse transcriptase polymerase chain reaction (RT-PCR) and immunocapture reverse transcriptase polymerase chain reaction (IC-RT-PCR) methods are available to distinguish LYSV from onion yellow dwarf virus in single and mixed infections (Dovas et al., 2001b; Lunello et al., 2005).



Figure 5. Adult and juvenile black bean aphids. Photo courtesy of Dr. Jim Hardie.

Known Distribution

Asia: Bangladesh, China, Japan, Indonesia, Iran, Thailand, Turkey, Vietnam, and Yemen. **Europe:** Belgium, Czech Republic, Denmark, Finland, France, Germany, Greece, Italy, Netherlands, and Sweden. **Oceania:** Australia and New Zealand. **North America:** Mexico and United States (Oregon, Washington). **South America:** Argentina, Brazil, Chile, Colombia, Uruguay, and Venezuela (Lunello et al., 2002; Pappu et al., 2005; Takaki et al., 2005; Perez-Moreno et al., 2006; Wei et al., 2006; CABI/EPPO, 2007; Gieck et al., 2007; Fidan and Baloglu, 2009a, b).

Potential Distribution within the United States

LYSV has been reported in garlic in Washington and Oregon (Pappu et al., 2005; Gieck et al., 2007). Although the virus is considered to be of restricted distribution in these states, the virus was found in the garlic germplasm maintained by the USDA-ARS Western Regional Plant Introduction Station in Washington (~15%) (Pappu et al., 2005; 2008). It was reported in 2006 that California led the way in garlic production in the United States, followed by Oregon and Nevada (Boriss, 2006). If LYSV travels south unchecked from where it has been reported in Oregon and Washington, it could spread to major commercial garlic producers in those states. Leek is not a common commercial product in the United States.

Pathway

LYSV can spread locally through aphid vectors (*M. persicae*, *A. fabae*) in a non-persistent manner, meaning that the virus does not replicate in the host, and stays localized on the

piercing stylet of the aphid. During feeding, the stylet breaks through plant tissue, and deposits the virus. Typically however, aphid vectors move the virus into the plot too late in the growing season to cause economic losses in that same season (Davis, 2008a).

Propagative plant material is the most important long-distance transmission pathway, since “the United States allows peeled garlic cloves from all countries without a permit...[and] imports *Allium* sp. for propagation from a wide variety of countries, including Mexico” (NPAG, 2009). The virus can also be transmitted in sap inoculation, a common practice in experimental settings.

Survey

CAPS-Approved Method*: The CAPS-approved survey method is to collect symptomatic plant tissue by visual survey.

Survey Sample Collection:

Leaves: Remove leaf tip samples from the second youngest leaf on symptomatic plants.

Bulbs: Remove bulb from the symptomatic plants of interest.

*For the most up-to-date methods for survey and identification, see Approved Methods on the CAPS Resource and Collaboration Site, at <http://caps.ceris.purdue.edu/>.

Literature-Based Methods:

In areas where the disease is known to occur it is recommended that at least 100 leaf samples (not necessarily symptomatic) per crop be collected if possible (Smith et al., 2006). From bulbs, remove one clove from the bulb, and cut a cube of approx. 1.0 x 0.5 x 0.5 cm from the basal region (Conci et al., 2002). The leaf tip and bulb cubes samples should be used in diagnostic analysis.

Key Diagnostics/Identification

CAPS-Approved Method*:

Enzyme Linked Immunosorbent Assay (ELISA): Agdia and AC diagnostics have commercially available double antibody sandwich (DAS) ELISAs for LYSV available using polyclonal antisera (https://orders.agdia.com/InventoryD.asp?attribute_Size=1000&collection=SRA+20301&loc=IN; <http://www.acdiainc.com/lysv.htm>). Neither of the commercial ELISA tests have been validated for regulatory purposes at this time, however. This test is to be used for screening only. Positive results will need to be verified using molecular methods.

*For the most up-to-date methods for survey and identification, see Approved Methods on the CAPS Resource and Collaboration Site, at <http://caps.ceris.purdue.edu/>.

Literature-Based Methods:

Indicator Hosts: (Bos, 1981)

- *Allium porrum*: Yellow leaf stripes are observed with infection of LYSV.

- *Chenopodium amaranticolor* and *C. quinoa*: Diagnostic symptoms of LYSV include chlorotic local lesions that become green rings when leaves senesce.
- *Celosia argentea*: brown necrotic local lesions are observed with infection of LYSV.

ELISA: Wei et al. (2006) extracted sap from the affected plant and tested for LYSV with general potyvirus monoclonal antibodies from Agdia Inc. (Elkhart, Indiana). They also used an ELISA with polyclonal antibodies (not known to be commercially available). The authors measured optical density of the ELISA plates at 405 nm. Positive readings are defined as any sample with an A_{405} of at least three times the mean of the negative control. Use healthy sap of non-host plants as the negative control. This diagnostic should be verified with RT-PCR or sequencing to confirm it is LYSV and not another potyvirus.

Immunoelectromicroscopy: Virus particles are trapped from crude leaf extracts on copper grids in 0.1 M phosphate buffer at pH 7.0 for 15 minutes. Grids are washed with 1:50 dilution of LYSV antiserum for 15 minutes. Decorated virus particles are then stained with 1% (weight/volume) of uranyl acetate (Dovas et al., 2001a).

Molecular:

RT-PCR: RT-PCR was performed using specific primers designed from the consensus regions of the coat protein genes of *Leek yellow stripe virus* (Fajardo et al., 2001). Viral RNA is extracted from purified virus samples or total RNA from infected plants. The cDNA is synthesized from the RNA using a “Time Saver cDNA Synthesis Kit” (Pharmacia Biotech). To amplify viral cDNA, add 10 μ L of the RT reaction to “50 μ L polymerase reaction mixture containing 20 mM Tris-HCl, pH 8.4, 50 mM KCl, 1.5 mM MgCl₂, 200 μ M each dNTPs, 2.5 U of Taq DNA polymerase” and 100 ng each of 1LYSV (5' TCA CTG CAT ATG CGC ACC AT 3') and 2LYSV (5' GCA CCA TAC AGT GAA TTG AG 3')” (Fajardo et al., 2001).

Run the reaction in a PCR protocol of “94°C for 5 min, followed by 35 cycles (94°C / 1 min, 50°C / 2 min and 72°C / 2 min) and a final amplification at 72°C for 7 min.” (Fajardo et al., 2001). The expected fragment is approximately 1000 base pairs (Fajardo et al., 2001). Pappu et al. (2008) also provide specific details on RT-PCR and primers for *Leek Yellow Stripe Virus*.



Figure 6. Diamond-shaped chlorotic lesions on onion seed stalks (top) and on onion leaves (bottom). Photos courtesy of Lindsey J. du Toit and Gary Q. Pelter.

Easily Confused Species

Leek Yellow Stripe Virus could be confused with *Iris Yellow Spot Virus* (IYSV) depending on the expression of symptoms (Pappu et al., 2005). Both of these viruses infect leek, but IYSV is a tospovirus, not a potyvirus. The main hosts of IYSV are onion and shallot, but the virus has the capacity to infect leek as well (Smith et al., 2006). IYSV is not known to infect any variety of garlic.

IYSV also causes chlorotic (yellow) lesions on unfurled leaves, but the lesions are distinctly diamond-shaped instead of continuous down the lamina as with LYSV (Fig. 6) (Schwartz et al., 2002). *Iris yellow spot virus* typically only infects leaves, compared to LYSV which infects bulb tissue with ease. IYSV is also established in many states, and is not considered an exotic pest (Smith et al., 2006).

IYSV vector transmission also differs from LYSV. *Iris yellow spot virus* is vectored by the onion thrips (*Thrips tabaci*) (Fig. 7) larvae in a persistent and propagative manner, meaning that the virus passes into the insect's salivary glands, and multiplies inside the vector. Transmission of IYSV therefore depends on the life stage of the vector and the success of replication inside that vector (Kritzman et al., 2001; Srinivasan et al., 2012).

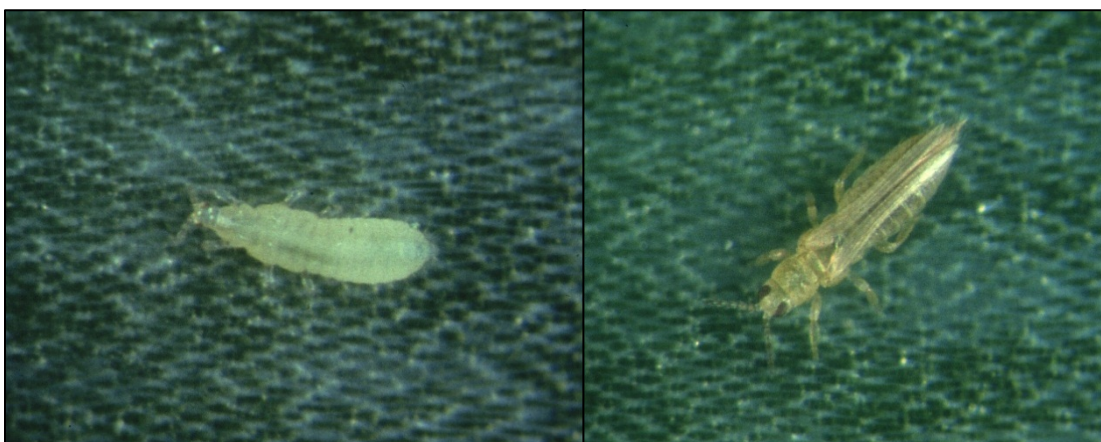


Figure 7. *Thrips tabaci* larvae (left) and adult (right). Photos courtesy of Joe Ogrodnick.

Another easily confused virus is *Onion Yellow Dwarf Virus* (OYDV). OYDV is also a potyvirus with similar symptoms to LYSV. Like LYSV, streaking starts at the base of the leaf and can spread to complete yellowing of the entire leaf. Leaves are sometimes flattened and fall over often. OYDV is transmitted by the same two species of aphids and via vegetative propagation, and is often found in complex with LYSV. The OYDV virus forms thread-like particles about 722 to 820 nm long (Davis, 2008b). To distinguish which virus is present in a symptomatic plant, it is best to use molecular diagnostics.

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This datasheet was developed by USDA-APHIS-PPQ-CPHST staff. Cite this document as:

Sullivan, M., and Robinson, A. 2012. CPHST Pest Datasheet for *Leek yellow stripe virus*. USDA-APHIS-PPQ-CPHST.