



Method for the detection of infectious tobamoviruses on tomato seed

Crop:	Tomato (<i>Lycopersicon esculentum</i> L. now <i>Solanum lycopersicum</i>)
Pathogen:	<i>Tobacco mosaic virus</i> (TMV) and <i>Tomato mosaic virus</i> (ToMV)
Revision history:	July 2017

Sample and sub-sample size

The recommended minimum sample size is 3,000 seeds with a maximum sub-sample size of 250 seeds.

Principle

- Detection of infectious tobamoviruses on tomato seed by using the local lesion assay; inoculation of indicator tobacco plant leaves with tomato seed ground in buffer. Infectious virions will cause a local lesion response (1). *Nicotiana tabacum* cv. *Xanthi* NN and/or *Nicotiana glutinosa* can be used as assay plants.

Restrictions on Use

- This test method is suitable for untreated seed.
- This test method is suitable for seed that has been treated using physical or chemical (acid extraction, calcium or sodium hypochlorite, tri-sodium phosphate, etc.) processes with the aim of disinfestation/disinfection, provided that any residue, if present, does not influence the assay. It is the responsibility of the user to check for such antagonism and/or inhibition by analysis, sample spiking, or experimental comparisons.
- This test method has not been validated for seed treated with protective chemicals or biological substances. If a user chooses to test treated seed using this method, it is the responsibility of the user to determine empirically (through analysis, sample spiking, or experimental comparisons) whether the protective chemicals or biological substances have an effect on the method results.

Validation

Results of a comparative test were approved by ISTA, see www.seedtest.org (>>Technical Committees >>Seed Health Committee >>Testing Methods >>Method Validation) and the method adopted as an ISTA Rule (7-028) in June 2012.

Some minor modifications proposed by ISHI-Veg were accepted by ISTA and the revised version of the method came into effect in January 2014.

This method has also been approved by the US National Seed Health System (NSHS) as a Standard B (see <http://seedhealth.org/seed-health-testing-methods/>).

Note: The method was reviewed recently and found to be fit for purpose. The section **Validation** has been updated and a section on **Method Execution** has been added.

Method Execution

To ensure process standardization and valid results, it is strongly recommended to follow the best practices developed by ISHI-Veg for *ELISA Assays in Seed Health Tests* (see <http://www.worldseed.org/our-work/phytosanitary-matters/seed-health/ishi-veg/>).

Method description

See www.seedtest.org (>>Technical Committees >>Seed Health Committee >>Testing Methods)