Method for the Detection of *Pseudomonas phaseolicola* pv. *phaseolicola* on Bean Seed

Crop:	Bean (*Phaseolus vulgaris* L.)
Pathogen:	*Pseudomonas syringae* pv. *phaseolicola* (Burkholder) Young et al. (*P. s.* pv. *phaseolicola*) (syn. *Pseudomonas savastanoi* pv. *phaseolicola*)
Date:	July 2017

Sample and sub-sample size

The test is done on a minimum sample size of 5,000 seeds and a maximum sub-sample size of 1,000 seeds.

Note: The method was validated using a minimum of 5,000 seeds. However, sample size depends on the risk management strategy of each user, and thus the choice of sample size is at the user's discretion.

Principle

- Detection of viable bacteria based on soaking seeds and plating the liquid obtained on semi selective media.
- Confirmation of suspect bacterial colonies is completed by a pathogenicity assay.

Restrictions on Use

- This test method is suitable for untreated seed.
- The ability to recover *P. s.* pv. *phaseolicola* on plates can be influenced by the presence of other microorganisms and/or inhibitory chemicals used for seed disinfestation/ disinfection. 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 validated by ISTA, see www.seedtest.org >>Technical Committees >>Seed Health Committee >>Testing Methods >>Method Validation). The method was adopted as an ISTA Rule (7-023) in January 2007.

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

Note: The method was reviewed recently and found to be fit for purpose. The sections **Sample and sub-sample size** and **Validation** have been updated. A new section **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 *Dilution Plating 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)