

ISTA Method Validation for Seed Testing

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The ISTA Method Validation Programme is:

“A critical examination of a seed quality test to ensure that:

- i) the description of the method is clear and complete
- ii) the procedure gives accurate, reproducible and repeatable results”

History

Pre-2000: Rules proposals varied from those which had undergone thorough collaborative studies to those which had not.

2000: Seed Health Committee produces “Handbook of Method Validation for the Detection of Seed-Borne Pathogens”; all Seed Health Rules proposals from that date have been validated.

2002: Seed Vigour Committee amends the Seed Health Handbook for use with Vigour Tests.

2002: ECOM decides that method validation should apply for all seed quality testing; Method Validation Working Group formed and charged with producing 'ISTA Method Validation for Seed Testing'; all methods already in the Rules are considered as validated.

2003-2005: Drafts one to four evolve;
Statistics Committee agrees to prepare an
Appendix on statistics for use in Method
Validation; ECOM requests inclusion of
information on validation of methods for
testing for the presence of specified traits.

2006: ECOM decides that

- i) starting with Rules Proposals 2008, methods submitted to the Ordinary Meeting must have been validated
- ii) the validated method itself must be included in the Rules Proposal document

Programme Guidelines

1. Collaboratively Validated Methods
 - a) Multi-laboratory validated test methods
 - b) Peer-validated test methods

2. Performance Validated Methods

Multi-Laboratory Validated Test Methods

- A multi-laboratory (minimum of six) collaborative study.
- Still the preferred type of method validation for ISTA.
- Little change from what many TCOMs have always done, except that reviewers are now required and a Method Validation Report must be produced and accepted by the TCOM before a Rules Proposal can be submitted.
- Hypothetical example: use of organic growing media in the germination test for *Phaseolus vulgaris*.

Peer-Validated Test Methods

- A minimum of two collaborating laboratories.
- A mechanism for “fast-tracking” method validation, but only applicable when the TCOM has decided a multi-laboratory collaborative study is not required.
- Reviewers and a Method Validation Report are still required.
- Hypothetical example: germination testing of a tree species by using counted replicates as well as the current weighed replicate method.

Performance Validated Methods

- A minimum of one laboratory.
- Verification of performance claims of the test method.
- Primarily for allowing the introduction of test kit methods into the Rules.
- Hypothetical example: test for endophyte (*Neotyphodium lolii*) in perennial ryegrass (*Lolium perenne*).

When will ISTA Method Validation be required?

1. For a test method where none currently exists in the Rules
e.g. Item 5a of the ISTA 2006 Rules Changes Proposals – “Detection of *Xanthomonas axonopodis* pv. *phaseoli* (common blight) on bean (*Phaseolus vulgaris*”. No method existed previously.

2. Where more than one test method is in common use, but none are currently in the Rules

e.g. accelerated ageing vigour testing for *Brassica* spp. has been proposed, but one group of laboratories uses 41°C/72h while another group of laboratories considers that 42°C/48h gives more consistent results (hypothetical example).

3. Where changing some aspect of a method already in the Rules will improve the method

e.g. Item 4a of the ISTA 2006 Rules Changes Proposals – “Modification to dormancy breaking for *Hordeum vulgare*”. Addition of KNO_3 provides a further option for breaking dormancy, thereby improving the method.

4. Replace an existing method in the Rules which has become outmoded by one which offers improved performance
e.g. replacing the agar plate test method for a seed-borne fungal pathogen by a method using molecular markers (hypothetical example).
5. Extend a test method to include a new species, or addition within a species
e.g. Item 6c of the ISTA 2006 Rules Changes Proposals – “Addition of *Crambe abyssinica*”. New species added to Table 5A.

In summary, method validation in ISTA is a five step process:

1. Method selection and development.
2. Validation through comparative testing.
3. Review of comparative test results and preparation of a Method Validation Report.
4. Approval of validation status by the TCOM and preparation of a Rules Proposal.
5. Acceptance by the ISTA membership and publication of the validated method in the Rules.