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International Rules for Seed Testing 2023

Chapter 7: Seed health testing

Including changes and editorial corrections adopted at the Ordinary General Meeting 2022, Cairo, Egypt

Effective from 1 January 2023

Note on the use of the translations

The electronic version of the International Rules for Seed Testing includes the English, French, German and Spanish versions. If there are any questions on interpretation of the ISTA Rules, the English version is the definitive version.

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Contents

Preface to the 2023 edition of the ISTA Rules

Since 2014, the *International Rules for Seed Testing* (ISTA Rules) are primarily available in electronic format. The ISTA Rules can be downloaded as a complete PDF file or as individual chapters from:

www.ingentaconnect.com/content/ista/rules.

If required, users of the ISTA Rules can print their own copies. For further information on the ISTA Rules, see:

www.seedtest.org/rules.

The electronic version includes the English, French, German and Spanish versions of the ISTA Rules. If there are any questions on interpretation of the ISTA Rules, the English version is the definitive version.

Seed health testing methods

Previously, the seed health testing methods were published as a separate Annexe to Chapter 7 of the ISTA Rules. They are now available as separate method sheets from the ISTA website at:

www.seedtest.org/seedhealthmethods.

Details of changes

The 2023 changes are editorial corrections or Rules changes adopted at the Ordinary General Meeting held at Cairo, Egypt in May 2022. Edits were made in Adobe InDesign by Vanessa Sutcliffe of HeartWood Editorial (www.heartwoodeditorial.co.uk).

The changes in the text content from the previous edition of the ISTA Rules are listed below. They can be displayed with yellow highlight boxes as a 'layer' over the English version within the electronic copy, with comments on what has changed.

For the previous history of amendments to the ISTA Rules, see the Prefaces for 2003 to 2022 on the ISTA website.

Ernest Allen, ISTA Rules Committee Chair

Susan Alvarez, ISTA Rules Committee Vice-Chair

ISTA Secretariat

Changes to the ISTA Rules for 2023

Chapter 1

1.1, 1.2, 1.2.4, 1.3, 1.4.1, 1.4.2, 1.4.3, 1.6: Edits made following offer of optional electronic seed analysis certificates from 2023, for use by accredited member laboratories of ISTA. Proposal developed by ISTA Rules Chair with input from Secretariat and ECOM.

Chapter 3

3.7: Paragraph moved for clarification of reporting authorities when determining scientific names for pure and other seeds reported on ISTA Certificates. Harmonisation of reporting between 3.7 and 4.7. Proposal developed by Purity TCOM and approved by unanimous vote.

3.7: Proposal developed and approved by Purity TCOM to indicate how other seeds and inert matter found in the second whole sample test shall be reported, providing clear guidance to the second whole sample test.

Chapter 4

4.7: Wording altered for clarification of reporting authorities when determining scientific names for pure and other seeds reported on ISTA Certificates. Harmonisation of reporting between 3.7 and 4.7. Proposal developed by Purity TCOM and approved by unanimous vote.

Chapter 5

5.6.2.3, 5.6.3.1: Proposal gives clarification that the temperature prescribed for prechilling should be measured from on or in the substrate during prechilling. Proposal developed and approved by Germination TCOM.

5.6.5.3: Proposal gives clarification that a tetrazolium test can be conducted to determine viability of fresh ungerminated seeds, at the end of a germination test, for species listed in Table 5A. Proposal developed by Germination TCOM and approved by Germination and Tetrazolium TCOMs.

5.10: Proposal to add Seedling Evaluation Groups to Table 5A (Parts 1, 2 and 3), giving benefit to seed analysts and ensuring that correct group is used. Proposal developed and approved by Germination TCOM.

Table 5A Part 1: Addition of new temperature regime (20 °C) for germination of *Anethum graveolens* following an ISTA peer method validation study. Proposal developed and approved by Germination TCOM.

Table 5A Parts 1, 2 and 3: Proposal to add Seedling Evaluation Groups to Table 5A, giving benefit to seed analysts and ensuring that correct group is used. Proposal developed and approved by Germination TCOM.

Chapter 7

Method 7-031: Addition of option to use a sieve with filter paper or an equivalent nematode-permeable container (such as a non-woven plant growth bag) for filtration. Proposal developed and approved by Seed Health TCOM, and supported by method validation study.

Chapter 8

8.10.4 (new section): Inclusion of DNA-based test for testing *Pisum* varieties. Proposal developed by working group within Variety TCOM and supported by validation study. Proposal approved by Variety TCOM.

8.10.5 (new section): Inclusion of DNA-based test for testing *Avena sativa*. Proposal supported by validation study. Proposal developed and approved by Variety TCOM.

Table 8I: Table relabelled as 8R due to addition of new methods in Chapter 8.

Chapter 9

9.2.7: Paragraph moved and bullet point added to improve clarity. Proposal developed and approved by Moisture TCOM.

9.3.2.6: Editorial correction to improve sentence clarity.

9.3.2.7: Bullet point added to improve clarity. Proposal developed and approved by Moisture TCOM.

Table 9A Part 1: Editorial corrections of nomenclature changes approved in 2019 *ISTA List of Stabilised Plant Names*. *Elytrigia* spp. updated to *Elymus* spp.

Chapter 19

19.1: Introduction of acronym TP for 'trait purity'. Small risk of ambiguity with TP used in Chapter 5 for 'top of paper'. In cases where ambiguity might exist, 'trait purity' should be used in full. Proposal developed and approved by GMO TCOM.

19.2: Editorial corrections made that do not modify intent, including renumbering of paragraphs and word substitutions.

19.2: New definitions added and numbering of definitions throughout section modified as a result.

19.2.17: Improvement of definition of term 'analyte', relating it to term 'target' widely used in Chapter 19.

19.2.20: Improved definition of 'performance-based approach' as a mechanism to ensure uniformity in testing.

19.2.21: Improved definition of 'proficiency test' to clarify object of assessing ability of subject (laboratory, operator) to carry out a test, not a specific method.

19.3: Section revised, including precise definition of pieces of evidence a laboratory must provide to apply for accreditation under performance-based approach. Requirement of method validation or verification is introduced. Precise specification of requirement of production of performance data on seed samples is given. Use by laboratories of results obtained in non-ISTA proficiency tests is accepted as evidence.

19.4: Section retitled 'Objectives and approaches' consistent with revised terminology. Testing objectives and approaches defined in relation to each other, with reference to Fig. 19.1. Two paragraphs describing 'technical aspects' moved to end of section. Sentence indicating need for reference material in quantitative PCR deleted since digital PCR does not necessarily require it. Several editorial changes made.

19.4.1: Redundant sentence deleted regarding size of working sample compared to submitted sample. Specification that working sample can be analysed in single or in multiple units of observation. Clarification of concept of limit of group/bulk size (not of working sample size) in relation to limit of quantification.

19.5: Section retitled 'Testing technologies' consistent with revised terminology.

19.5.1.1: Minor changes and specifications in bullet points.

19.5.1.3: Addition of need to verify identity of PCR products in real-time PCR when using intercalating dyes.

19.5.2.2: Simplification of text on lateral flow strip test.

19.5.2.3: Simplification of text on enzyme-linked immunosorbent assay.

19.5.3.1: Replacement of 'resistance' with 'tolerance' as a more appropriate term when referring to herbicides.

19.6.1: Specification added that testing results for both assessment of presence or estimation of level can be referred either to seed sample or seed lot. Deletion of unnecessary sentence, and other minor changes and specifications.

19.6.2: Modification of section title and text for consistency with revised terminology and to improve clarity.

19.6.3: New reference (Remund *et al*., 2020) added.

19.7: Specification of details to be provided when reporting results on an ISTA Certificate, including conditions for reporting results of GMO testing on Orange or Blue International Certificates.

19.7.1: Section retitled 'Assessment of presence of GMO' consistent with revised terminology, and editorial changes made. Clarification of how to express compliance to a given standard when a working sample tests negative to a qualitative assay for a given target.

19.7.2: Section retitled 'Estimation of the level of GMO by multiple qualitative tests of individuals or groups' consistent with revised terminology.

19.7.3: Section retitled 'Estimation of the level of GMO by quantitative measurements on groups or bulks' consistent with revised terminology. Specification of conditions for reporting results of GMO testing on Orange or Blue International Certificates. Other editorial changes made to improve clarity.

19.8: New reference (Remund *et al*., 2020) added and link for SeedCalc download on ISTA website updated.

Table 19A (new table): Table to summarise types of analytical output that can be attained using different approaches in GMO testing.

Figure 19.1: Figure redrawn according to revised terminology. More information provided including new concepts (unit of observation, testing approach) and relating workflow to different analytical outputs.

Chapter 7: Seed health testing

7.1 Object

The object of a seed health test is to determine the health status of a seed sample, and by inference that of the seed lot.

Health testing of seed is important for four reasons:

a. Seed-borne inoculum may give rise to progressive disease development in the field and reduce the commercial value of the crop.
b. Imported seed lots may introduce diseases into new regions. Tests to meet quarantine requirements may therefore be necessary.
c. Seed health testing may elucidate seedling evaluation and causes of poor germination or field establishment and thus supplement germination testing.
d. Seed health test results can/may indicate the necessity to carry out/perform seed lot treatment(s) in order to eradicate seed-borne pathogens or to reduce the risk of disease transmission.

7.2 Definitions

7.2.1 Seed health

Health of seed refers primarily to the presence or absence of disease-causing organisms, such as fungi, bacteria and viruses, and animal pests, including nematodes and insects, but physiological conditions such as trace element deficiency may be involved.

7.2.2 Pretreatment

Any physical or chemical laboratory treatment of the working sample preceding incubation, given solely to facilitate testing.

7.2.3 Seed treatment

See 2.2.12. For seed health testing, a seed lot may be treated for the purpose of controlling plant pathogens or insect pests, or correcting trace element deficiencies.

7.2.4 ISTA Seed Health Method Validation Programme

Before publication in the *International Rules for Seed Testing*, the ISTA seed health testing methods (new or equivalent) are validated. The principles and factors which should be considered in the validation of methods for the detection of seed-borne pathogens are described in the *ISTA Technical Guidelines for Organising and Analysing Results of Proficiency Tests (PT) and Interlaboratory Tests for Validation of Methods (CT)*.

7.3 General principles

Seed health testing should be performed using methods and equipment which have been tested to ensure they are fit for purpose. Different methods of testing are available, varying in sensitivity and reproducibility and in the amount of training and equipment required. The method used will depend on the pathogen or condition to be investigated, the species of the seed, and the purpose of the test. Selection of the method and evaluation of the results requires knowledge and experience of the methods available. The presence or absence of disease organisms, pests and deleterious physiological conditions specified by the sender is estimated as accurately as the method used permits.

7.4 Procedures

7.4.1 Working sample

The entire submitted sample, or a proportion of it, depending on the test method, may be used as a working sample. The sample should be packaged and submitted in a manner which will not alter its seed health status.

Exceptionally, a submitted sample larger than that prescribed in 2.8 may be required and in such cases the sampler must be instructed accordingly.

When a portion of the submitted sample is required as a working sample, the reduction must be carried out in accordance with 2.5.2, taking appropriate precautions to avoid cross-contamination.

Normally the working sample must not be less than that specified in the method description.

Replicates containing a specified number of seeds, if required, must be taken at random from a subsample after thorough mixing.

7.4.2 Seed treatment

Test results may be influenced by treatment applied to the seed lot. Seed health tests on treated seeds will generally deliver unreliable test results caused by masking or inhibition of the growth of the target organism. Individual Method Sheets will determine whether the testing of treated seeds is acceptable.

7.4.3 Sample storage

The microflora of seed, in the lot or the sample, may change considerably during storage in conditions in which seed viability is satisfactorily maintained. The selection of the appropriate storage conditions must take into account the optimal storage temperature and container in order to maintain sample integrity.

Abundant development of saprophytic moulds including 'storage fungi' in tests can be an indication that the seed is not of good quality due to unfavourable harvesting, processing or storage conditions, or to ageing. Some fungi (such as *Rhizopus* spp.) spread rapidly over tests on blotters and may rot originally healthy seedlings or may interfere with outgrowth of the pathogen from the plated infected seeds. Pretreatment as described in the specific method may be advisable.

7.4.4 Specific directions

Specific seed health testing methods are published online on the ISTA web site at:

www.seedtest.org/seedhealthmethods

Seed health methods are normally based on one host, and one pathogen, but multi-pathogen methods may be included. Before publication, all seed health test methods must be validated through the ISTA Seed Health Method Validation Programme. Methods validated in this way at the time of printing are listed in Table 7A. Additions, updates and deletions to this list can be found on the ISTA web site (www.seedtest.org/seedhealthmethods). The definitive list is held by the ISTA Secretariat. It is the responsibility of the laboratory using the method to consult this list.

7.5 Calculation and expression of results

Results are expressed either qualitatively or quantitatively as specified in the individual prescribed methods.

7.6 Reporting results

The results of a test for seed health must be reported under 'Other determinations' as follows:

- either qualitative or quantitative results, as specified in the individual methods;
- negative and positive results, as specified in the individual methods;
- the scientific name of the pathogen detected;
- the percentage of infected seeds;
- the method used, including any pretreatment (7.2.2);
- the size of the sample or fraction examined;
- any additional permitted procedure used.

The absence of a statement concerning the health condition of the seed does not necessarily imply that the health condition is satisfactory.

Table 7A. ISTA official seed health testing methods

7-001a: Detection of *Alternaria dauci* in *Daucus carota* (carrot) seed by blotter method
Host: *Daucus carota* L.
Pathogen(s): *Alternaria dauci* (J.G.Kühn) J.J.Groves & Skolko, syn. *A. porri* f.sp. *dauci* (J.G.Kühn) Neerg., syn. *A. carotae* (Ellis & Langlois) Stevenson & Wellman
Date approved: 2012
Review due: 2017

7-001b: Detection of *Alternaria dauci* in *Daucus carota* (carrot) seed by malt agar method
Host: *Daucus carota* L.
Pathogen(s): *Alternaria dauci* (J.G.Kühn) J.J.Groves & Skolko, syn. *A. porri* f.sp. *dauci* (J.G.Kühn) Neerg., syn. *A. carotae* (Ellis & Langlois) Stevenson & Wellman
Date approved: 2012
Review due: 2017

7-002a: Detection of *Alternaria radicina* in *Daucus carota* (carrot) seed by blotter method
Host: *Daucus carota* L.
Pathogen(s): *Alternaria radicina* Meier, Drechsler & E.D.Eddy, syn. *Stemphylium radicinum* (Meier, Drechsler & E.D.Eddy) Neergaard
Date approved: 2012
Review due: 2017

7-002b: Detection of *Alternaria radicina* in *Daucus carota* (carrot) seed by malt agar method
Host: *Daucus carota* L.
Pathogen(s): *Alternaria radicina* Meier, Drechsler & E.D.Eddy, syn. *Stemphylium radicinum* (Meier, Drechsler & E.D.Eddy) Neergaard
Date approved: 2012
Review due: 2017

7-003: Detection of *Botrytis cinerea* in *Helianthus annuus* (sunflower) seed
Host: *Helianthus annuus* L.
Pathogen(s): *Botrytis cinerea* Pers. ex Pers. (Perfect state *Botryotinia fuckeliana* (de Bary) Whetzel, syn. *Sclerotinia fuckeliana* (de Bary) Fuckel.)
Date approved: 2011
Review due: 2016

7-004: Detection of *Leptosphaeria maculans* and *Plenodomus biglobosus* in *Brassica* spp. seed
Host: *Brassica* spp.
Pathogen(s): *Leptosphaeria maculans* (Tode ex Fr.) Ces. & de Not (previously *Phoma lingam*) or *Plenodomus biglobosus* (Shoemaker & H. Brun) (previously *Leptosphaeria biglobosa*)
Date approved: 2017
Review due: 2022

7-005: Detection of *Ascochyta pisi* in *Pisum sativum* (pea) seed
Host: *Pisum sativum* L.s.l.
Pathogen(s): *Ascochyta pisi* Lib.
Date approved: 2022
Review due: 2027

7-006: Detection of *Colletotrichum lindemuthianum* in *Phaseolus vulgaris* (bean) seed
Host: *Phaseolus vulgaris* L.
Pathogen(s): *Colletotrichum lindemuthianum* (Sacc. & Magn.) Briosi & Cav.
Date approved: 2022
Review due: 2027

7-007: Detection of *Alternaria linicola*, *Botrytis cinerea* and *Colletotrichum lini* in *Linum usitatissimum* (flax, linseed) seed
Host: *Linum usitatissimum* L.
Pathogen(s): *Alternaria linicola* J.W.Groves & Skolko; *Botrytis cinerea* Pers. ex Pers. (Perfect state *Botryotinia fuckeliana* (de Bary) Whetzel, syn. *Sclerotinia fuckeliana* (de Bary) Fuckel.); *Colletotrichum lini* (Westerd.) Tochinai, syn. *C. linicola* Pethybr. & Laff.
Date approved: 2012
Review due: 2017

7-008: Detection of *Caloscypha fulgens* in *Picea engelmannii* and *P. glauca* (spruce) seed
Host: *Picea engelmannii* Engelm.; *Picea glauca* (Moench) Voss
Pathogen(s): *Caloscypha fulgens* (Pers.) Boud. (Imperfect state *Geniculodendron pyriforme* Salt)
Date approved: 2011
Review due: 2016

Table 7A. ISTA official seed health testing methods (cont.)

7-009: Detection of *Fusarium circinatum* 1n *Pinus* spp. (pine) and *Pseudotsuga menziesii* (Douglas fir) seed
Host: *Pinus* spp.; *Pseudotsuga menziesii* (Mirb.) Franco
Pathogen(s): *Fusarium circinatum* Nirenberg & O'Donnell (syn. *Fusarium subglutinans* f. sp. *pini* Hepting, syn. *Fusarium lateritium* f. sp. *pini* Hepting, syn. *Gibberella circinata*)
Date approved: 2018
Review due: 2023

7-010: Detection of *Bipolaris oryzae* in *Oryza sativa* (rice) seed
Host: *Oryza sativa* L.
Pathogen(s): *Bipolaris oryzae* (Breda de Haan) Shoem., syn. *Drechslera oryzae*, syn. *Helminthosporium oryzae* Breda de Haan (Perfect state *Cochliobolus miyabeanus* (Ito & Kurib.) Drechsler ex Dastur, syn. *Ophiobolus miyabeanus* Ito & Kuribayashi)
Date approved: 2018
Review due: 2023

7-011: Detection of *Pyricularia oryzae* in *Oryza sativa* (rice) seed
Host: *Oryza sativa* L.
Pathogen(s): *Magnaporthe grisea* (Hebert) Barr (Imperfect state *Pyricularia oryzae* Cavara, syn. *P. grisea*)
Date approved: 2011
Review due: 2016

7-012: Detection of *Trichoconiella padwickii* in *Oryza sativa* (rice) seed
Host: *Oryza sativa* L.
Pathogen(s): *Trichoconiella padwickii* Ganguly, syn. *Alternaria padwickii* (Ganguly) Jain
Date approved: 2018
Review due: 2023

7-013a: Detection of *Ustilago nuda* in *Hordeum vulgare* subsp. *vulgare* (barley) seed by embryo extraction
Host: *Hordeum vulgare* L. subsp. *vulgare*
Pathogen(s): *Ustilago nuda* (Jens.) Rostr.
Date approved: 2022
Review due: 2027

7-013b: Detection of *Ustilago nuda* in *Hordeum vulgare* subsp. *vulgare* (barley) seed by dehulling and embryo extraction
Host: *Hordeum vulgare* L. subsp. *vulgare*
Pathogen(s): *Ustilago nuda* (Jens.) Rostr.
Date approved: 2022
Review due: 2027

7-014: Detection of *Parastagonospora nodorum* in *Triticum aestivum* subsp. *aestivum* (wheat) seed
Host: *Triticum aestivum* L. subsp. *aestivum*
Pathogen(s): *Parastagonospora nodorum* (Berk.) Quaedvl., Verkley & Crous 2013, syn. *Stagonospora nodorum*, syn. *Septoria nodorum* Berk. (Perfect state *Leptosphaeria nodorum* Mailer)
Date approved: 2022
Review due: 2027

7-015: Detection of *Epichloë coenophiala* in *Festuca* spp. (fescue) and of *Neotyphodium lolii* in *Lolium* spp. (ryegrass) seed
Host: *Festuca* spp., *Lolium* spp.
Pathogen(s): *Epichloë coenophiala* (Morgan-Jones & W. Gams) C.W. Bacon & Schardl; *Neotyphodium lolii* (Latch, M.J.Chr. & Samuels) Glenn, C.W.Bacon & Hanlin
Date approved: 2017
Review due: 2022

7-016: Detection of *Phomopsis* complex in *Glycine max* (soybean, soya bean) seed
Host: *Glycine max* (L.) Merr.
Pathogen(s): *Phomopsis longicolla* Hobbs, *Diaporthe phaseolorum* var. *sojae* (Lehm.) Wehm. (Imperfect state *P. phaseoli* (Desm.) Sacc., syn. *P. sojae* Lehmann); *Diaporthe phaseolorum* (Cke. & Ell.) Sacc. f. sp. *caulivora* (DPC), syn. *D. phaseolorum* var. *caulivora* Athow & Caldwell
Date approved: 2022
Review due: 2027

7-017: (Replaced by 7-007)

7-018: (Replaced by 7-007)

7-019a: Detection of *Xanthomonas campestris* pv. *campestris* and *Xanthomonas campestris* pv. *raphani* in *Brassica* spp. seed
Host: *Brassica* spp.
Pathogen(s): *Xanthomonas campestris* pv. *campestris* (Pammel) Dowson and *Xanthomonas campestris* pv. *raphani*
Date approved: 2022
Review due: 2027

7-019b: Detection of *Xanthomonas campestris* pv. *campestris* in disinfested/disinfected *Brassica* spp. seed
Host: *Brassica* spp.
Pathogen(s): *Xanthomonas campestris* pv. *campestris* (Pammel) Dowson
Date approved: 2018
Review due: 2023

Table 7A. ISTA official seed health testing methods (cont.)

7-020: Detection of *Xanthomonas hortorum* pv. *carotae* in *Daucus carota* (carrot) seed
Host: *Daucus carota* L.
Pathogen(s): *Xanthomonas hortorum* pv. *carotae* (Kendrick) Vauterin, Hoste, Kersters & Swings, syn. *X. campestris* pv. *carotae* (Kend) Dye
Date approved: 2010
Review due: 2015

7-021: Detection of *Xanthomonas axonopodis* pv. *phaseoli* and *X. axonopodis* pv. *phaseoli* var. *fuscans* in *Phaseolus vulgaris* (bean) seed
Host: *Phaseolus vulgaris* L.
Pathogen(s): *Xanthomonas axonopodis* pv. *phaseoli* (Smith) Vauterin, Hoste, Kersters & Swings, syn. *X. campestris* pv. *phaseoli* (Smith) Dye; *Xanthomonas axonopodis* pv. *phaseoli* var. *fuscans* Vauterin, Hoste, Kersters & Swings, syn. *X. campestris* pv. *phaseoli* var. *fuscans* (Burkholder) Starr & Burkholder
Date approved: 2011
Review due: 2016

7-022: Detection of *Microdochium nivale* and *M. majus* in *Triticum* spp. (wheat) seed
Host: *Triticum* spp.
Pathogen(s): *Microdochium nivale* Samuels & Hallett, syn. *Fusarium nivale* (Fr.) Rabenh. (Perfect state *Monographella nivalis* (Schaff.) Müller); *M. majus* (Wollenw.) Glynn & S.G.Edwards, syn. *M. nivale* var. *majus* (Wollenw.) Samuels & I.C.Hallett
Date approved: 2022
Review due: 2027

7-023: Detection of *Pseudomonas savastanoi* pv. *phaseolicola* in *Phaseolus vulgaris* (bean) seed
Host: *Phaseolus vulgaris* L.
Pathogen(s): *Pseudomonas savastanoi* pv. *phaseolicola* (Burkh.) Gardan, Bollet, Abu, Ghorrah, Grimont & Grimont, syn. *P. syringae* pv. *phaseolicola* (Burkh.) Young, Dye & Wilkie
Date approved: 2012
Review due: 2017

7-024: Detection of *pea early browning virus* and *pea seed-borne mosaic virus* in *Pisum sativum* (pea) seed
Host: *Pisum sativum* L.s.l.
Pathogen(s): *Pea early browning virus* (PEBV) and *pea seed-borne mosaic virus* (PSbMV)
Date approved: 2012
Review due: 2017

7-025: Detection of *Aphelenchoides besseyi* in *Oryza sativa* (rice) seed
Host: *Oryza sativa* L.
Pathogen(s): *Aphelenchoides besseyi* Christie
Date approved: 2019
Review due: 2024

7-026: Detection of *squash mosaic virus*, *cucumber green mottle mosaic virus* and *melon necrotic spot virus* in cucurbit seed
Host: Cucurbits
Pathogen(s): *Squash mosaic virus* (SqMV); *cucumber green mottle mosaic virus* (CGMMV); *melon necrotic spot virus* (MNSV)
Date approved: 2014
Review due: 2019

7-027: Detection of *Pyrenophora teres* and *P. graminea* in *Hordeum vulgare* subsp. *vulgare* (barley) seed
Host: *Hordeum vulgare* L. subsp. *vulgare*
Pathogen(s): *Pyrenophora teres* Drechsler (Imperfect state *Drechslera teres* (Sacc.) Shoem.); *Pyrenophora graminea* Ito & Kurib. (Imperfect state *D. graminea* (Rabenh. Ex Schlecht.) Shoem.)
Date approved: 2011
Review due: 2016

7-028: Detection of infectious *tobacco mosaic virus* and *tomato mosaic virus* in *Solanum lycopersicum* (tomato) seed by the local lesion assay (indexing) in *Nicotiana tabacum* plants
Host: *Solanum lycopersicum* L.
Pathogen(s): *Tobacco mosaic virus* (TMV); *tomato mosaic virus* (ToMV)
Date approved: 2012
Review due: 2017

7-029: Detection of *Pseudomonas syringae* pv. *pisi* in *Pisum sativum* (pea) seed
Host: *Pisum sativum* L.s.l.
Pathogen(s): *Pseudomonas syringae* pv. *pisi* (Sack.) Young, Dye & Wilkie
Date approved: 2012
Review due: 2017

7-030: Detection of *Acidovorax valerianellae* in *Valerianella locusta* (corn salad) seed
Host: *Valerianella locusta* (L.) Laterr.
Pathogen(s): *Acidovorax valerianellae* sp. nov.
Date approved: 2014
Review due: 2019

Table 7A. ISTA official seed health testing methods (cont.)

7-031: Filtration method for detection of *Ditylenchus dipsaci* in *Medicago sativa*; *D. dipsaci* and *D. gigas* in *Vicia faba*
Host: *Medicago sativa* L. and *Vicia faba* L.
Pathogen(s): *Ditylenchus dipsaci* Kuhn, 1857; *Ditylenchus gigas* n. sp.
Date approved: 2023
Review due: 2028

7-032: Detection of *Verticillium dahliae* in *Spinacia oleracea* (spinach) seed
Host: *Spinacia oleracea* L.
Pathogen(s): *Verticillium dahliae* Kleb.
Date approved: 2017
Review due: 2022