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| International Union for the Protection of New Varieties of Plants |  |

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| Technical Committee  Fifty-Fifth Session Geneva, October 28 and 29, 2019 | TC/55/23  Original: English  Date: October 11, 2019 |

Partial revision of the Test Guidelines for TOMATO ROOTSTOCKS

Document prepared by experts from the Netherlands

Disclaimer: this document does not represent UPOV policies or guidance

The purpose of this document is to present a proposal for a partial revision of the Test Guidelines for Tomato rootstocks (document TG/294/1 Corr. Rev. 2).

The TWV, at its fifty-third session, held in Seoul, Republic of Korea, from May 20 to 24, 2019, considered a proposal for a partial revision of the Test Guidelines for Tomato Rootstocks on the basis of documents TG/294/1 Corr. Rev. 2 and TWV/53/7 “Partial revision of the Test Guidelines for Tomato Rootstocks” and proposed the following changes (see document TWV/53/14 Rev. “Revised Report”, paragraph 95):

1. To change the denomination of the races of Characteristics 24.1, 24.2 and 24.3 “Resistance to *Fusarium oxysporum* f. sp. *lycopersici* (Fol)”;
2. To change the explanation Ad. 24 in Chapter 8.2 “Explanations for individual characteristics”;
3. To change the example varieties of Characteristic 28 “Resistance to *Pyrenochaeta lycopersici* (Pl)”;
4. To change the explanation Ad. 28 in Chapter 8.2 “Explanations for individual characteristics”

The proposed changes are presented from page 2 in highlight and underline (insertion) and ~~strikethrough~~ (deletion).

The TWV further agreed that the e-mail addresses below for obtaining the inoculum in all disease resistance explanations throughout the Test Guidelines for Tomato Rootstocks should be updated in the final adopted version of this partial revision of the Test Guidelines for Tomato Rootstocks as follows (see document TWV/53/14 Rev. “Revised Report”, paragraph 96):

Geves: matref@geves.fr

Naktuinbouw: resistentie@naktuinbouw.nl

INIA: resistencias@inia.sp

Proposal to change the denomination of the races of Characteristics 24.1, 24.2 and 24.3 “Resistance to *Fusarium oxysporum* f. sp. *lycopersici* (Fol)”

*Current wording*

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
|  |  | English | français | deutsch | español | Example Varieties Exemples Beispielssorten Variedades ejemplo | Note/ Nota |
| 24.  (+) |  | Resistance to  *Fusarium oxysporum* f. sp. *lycopersici* (Fol) | Résistance à *Fusarium oxysporum* f. sp. *lycopersici* (Fol) | Resistenz gegen *Fusarium oxysporum* f. sp. *lycopersici* (Fol) | Resistencia a  *Fusarium oxysporum* f. sp. *lycopersici* (Fol) |  |  |
| 24.1 (\*) | VG | – Race 0 (ex 1) | – Pathotype 0 (ex 1) | – Pathotyp 0 (ex 1) | – Raza 0 (ex 1) |  |  |
| **QL** |  | absent | absente | fehlend | ausente |  | 1 |
|  |  | present | présente | vorhanden | presente | Emperador | 9 |
| 24.2 (\*) | VG | – Race 1 (ex 2) | – Pathotype 1 (ex 2) | – Pathotyp 1 (ex 2) | – Raza 1 (ex 2) |  |  |
| **QL** |  | absent | absente | fehlend | ausente |  | 1 |
|  |  | present | présente | vorhanden | presente | Emperador | 9 |
| 24.3 (\*) | VG | – Race 2 (ex 3) | – Pathotype 2 (ex 3) | – Pathotyp 2 (ex 3) | – Raza 2 (ex 3) |  |  |
| **QL** |  | absent | absente | fehlend | ausente | Emperador | 1 |
|  |  | present | présente | vorhanden | presente | Colosus | 9 |

*Proposed new wording*

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
|  |  | English | français | deutsch | español | Example Varieties Exemples Beispielssorten Variedades ejemplo | Note/ Nota |
| 24.  (+) |  | Resistance to  *Fusarium oxysporum* f. sp. *lycopersici* (Fol) | Résistance à *Fusarium oxysporum* f. sp. *lycopersici* (Fol) | Resistenz gegen *Fusarium oxysporum* f. sp. *lycopersici* (Fol) | Resistencia a  *Fusarium oxysporum* f. sp. *lycopersici* (Fol) |  |  |
| 24.1 (\*) | VG | **– Race ~~0 (ex 1)~~ 0EU/1US** | **– Pathotype ~~0 (ex 1)~~ 0EU/1US** | **– Pathotyp ~~0 (ex 1)~~ 0EU/1US** | **– Raza ~~0 (ex 1)~~ 0EU/1US** |  |  |
| **QL** |  | absent | absente | fehlend | ausente |  | 1 |
|  |  | present | présente | vorhanden | presente | Emperador | 9 |
| 24.2 (\*) | VG | **– Race ~~1 (ex 2)~~ 1EU/2US** | **– Pathotype ~~1 (ex 2)~~ 1EU/2US** | **– Pathotyp ~~1 (ex 2)~~ 1EU/2US** | **– Raza ~~1 (ex 2)~~ 1EU/2US** |  |  |
| **QL** |  | absent | absente | fehlend | ausente |  | 1 |
|  |  | present | présente | vorhanden | presente | Emperador | 9 |
| 24.3 (\*) | VG | **– Race ~~2 (ex 3)~~ 2EU/3US** | **– Pathotype ~~2 (ex 3)~~ 2EU/3US** | **– Pathotyp ~~2 (ex 3)~~ 2EU/3US** | **– Raza ~~2 (ex 3)~~ 2EU/3US** |  |  |
| **QL** |  | absent | absente | fehlend | ausente | Emperador | 1 |
|  |  | present | présente | vorhanden | presente | Colosus | 9 |

Proposal to change the explanation Ad. 24 in Chapter 8.2 “Explanations for individual characteristics”

*Current wording*

Ad. 24: Resistance to *Fusarium oxysporum* f. sp. *lycopersici* (Fol)

1. Pathogen *Fusarium oxysporum* f. sp. *lycopersici*

3. Host species *Solanum lycopersicum*

4. Source of inoculum Naktuinbouw[[1]](#footnote-2) (NL) and GEVES[[2]](#footnote-3) (FR)

5. Isolate Race 0 (ex 1) (e.g. strains Orange 71 or PRI 20698 or Fol 071 1

(ex 2) (e.g. strains 4152 or PRI40698 or RAF 70 and 2 (ex 3)

Individual strains may vary in pathogenicity

6. Establishment isolate identity use differential varieties (see 9.3)

7. Establishment pathogenicity on susceptible tomato varieties

8. Multiplication inoculum

8.1 Multiplication medium Potato Dextrose Agar, Medium “S” of Messiaen

8.4 Inoculation medium………………….... water for scraping agar plates or Czapek-Dox culture medium (7 d‑old aerated culture)

8.6 Harvest of inoculum filter through double muslin cloth

8.7 Check of harvested inoculum spore count; adjust to 106 per ml

8.8 Shelf-life/viability inoculum 4-8 h, keep cool to prevent spore germination

9. Format of the test

9.1 Number of plants per genotype at least 20 plants

9.2 Number of replicates……………… 1 replicate

9.3 Control varieties for the test with race 0 (ex 1)

Susceptible………………………………(*Solanum lycopersicum*) Marmande, Marmande verte, Resal

Resistant for race 0 only……………….(*Solanum lycopersicum*) Marporum, Larissa, “Marporum x Marmande verte”, Marsol, Anabel

Resistant for race 0 and 1 ……….……..(*Solanum lycopersicum*) Motelle, Gourmet, Mohawk

Control varieties for the test with race 1 (ex 2)

Susceptible ………………………(*Solanum lycopersicum*) Marmande verte, Cherry Belle, Roma

Resistant for race 0 only ……………..(*Solanum lycopersicum*) Marporum, Ranco

Resistant for race 0 and 1 ………….…(*Solanum lycopersicum*) Tradiro, Odisea

Remark: ……….Ranco is slightly less resistant than Tradiro

Control varieties for the test with race 2 (ex 3)

Susceptible for race 2……………………Emperador

Resistant for race 0, 1 and 2…….………Colosus

9.4 Test design………………………… >20 plants; e.g. 35 seeds for 24 plants, including 2 blanks

9.5 Test facility ………………………… glasshouse or climate room

9.6 Temperature………………… 24-28°C (severe test, with mild isolate)

20-24°C (mild test, with severe isolate)

9.7 Light………………………………… 12 hours per day or longer

9.8 Season ………………………… all seasons

9.9 Special measures………………… slightly acidic peat soil is optimal; keep soil humid but avoid water stress

10. Inoculation

10.1 Preparation inoculums…………… aerated Messiaen or PDA or Agar Medium S of Messiaen or Czapek Dox culture or scraping of plates

10.2 Quantification inoculums………… spore count, adjust to 106 spores per ml,

Lower concentration for a very aggressive isolate

10.3 Plant stage at inoculation………… 10-18 d, cotyledon to first leaf

10.4 Inoculation method………………… roots and hypocotyls are immersed in spore suspension

for 5-15 min; trimming of roots is an option

10.7 Final observations………………… 14-21 days after inoculation

11. Observations

11.1 Method ………………………… visual

11.2 Observation scale………………… Symptoms:

growth retardation, wilting, yellowing,

vessel browning extending above cotyledon

11.3 Validation of test…………………… evaluation of variety resistance should be calibrated with results of resistant and susceptible controls

12. Interpretation of test results in comparison with control varieties

absent ………………………… [1] severe symptoms

present………………………… [9] mild or no symptoms

13. Critical control points:

Test results may vary slightly in inoculum pressure due to differences in isolate, spore concentration, soil humidity and temperature. Standards near borderline R/S will help to compare between labs.

*Proposed new wording*

Ad. 24: Resistance to *Fusarium oxysporum* f. sp. *lycopersici* (Fol)

|  |  |  |
| --- | --- | --- |
| 1. | Pathogen | *Fusarium oxysporum* f. sp. *lycopersici* |
| 3. | Host species | *Solanum lycopersicum* |
| 4. | Source of inoculum | Naktuinbouw[[3]](#footnote-4) (NL), GEVES[[4]](#footnote-5) (FR) or INIA[[5]](#footnote-6) (ES) |
| 5. | Isolate | race ~~0 (ex 1)~~0EU/1US(e.g. strains Orange 71 or PRI 20698 or Fol 071)  race ~~1 (ex 2)~~1EU/2US(e.g. strains 4152 or PRI40698 or RAF 70)  race ~~2 (ex 3)~~2EU/3US (e.g. strain Fol029)  ~~individual strains may vary in pathogenicity~~ |
| 6. | Establishment isolate identity | use differential varieties (see 9.3) |
| 7. | Establishment pathogenicity | on susceptible tomato varieties |
| 8. | Multiplication inoculum |  |
| 8.1 | Multiplication medium | Potato Dextrose Agar, Medium “S” of Messiaen |
| 8.4 | Inoculation medium | water for scraping agar plates or Czapek-Dox culture medium (7 d-old aerated culture) |
| 8.6 | Harvest of inoculum | filter through double muslin cloth |
| 8.7 | Check of harvested inoculum | spore count; adjust to 106 per ml |
| 8.8 | Shelflife/viability inoculum | 4-8 h, keep cool to prevent spore germination |
| 9. | Format of the test |  |
| 9.1 | Number of plants per genotype | at least 20 plants |
| 9.2 | Number of replicates | 1 replicate |
| 9.3.1 | Control varieties for the test with race ~~0 (ex 1)~~ 0EU/1US |  |
|  | Susceptible | (*Solanum lycopersicum*) Marmande, Marmande verte, Resal |
|  | Resistant ~~for race 0 only~~ | Emperador, Colosus and (*Solanum lycopersicum*) “Marporum x Marmande verte”, ~~Marsol, Anabel~~ Motelle, Gourmet, Mohawk, Ranco, Tradiro |
|  | ~~Resistant for race 0 and 1~~ | ~~(~~*~~Solanum lycopersicum~~*~~) Motelle, Gourmet, Mohawk~~ |
| 9.3.2 | Control varieties for the test with race ~~1 (ex 2)~~1EU/2US |  |
|  | Susceptible | (*Solanum lycopersicum*) Marmande verte, Cherry Belle, Roma, Marporum, Ranco |
|  | ~~Resistant for race 0 only~~ | ~~(~~*~~Solanum lycopersicum~~*~~) Marporum, Ranco~~ |
|  | Resistant ~~for race 0 and 1~~ | Emperador, Colosus and (*Solanum lycopersicum*) Tradiro, Odisea, “Motelle x Marmande verte” |
| 9.3.3 | Control varieties for the test with  race ~~2 (ex 3)~~2EU/3US |  |
|  | Susceptible ~~for race 2~~ | Emperador and (*Solanum lycopersicum*) Marmande verte, Motelle, Marporum |
|  | Resistant ~~for race 0, 1 and 2~~ | Colosus and (*Solanum lycopersicum*) Tributes, Murdoch, “Marmande verte x Florida” |
| 9.4 | Test design | >20 plants; e.g. 35 seeds for 24 plants, including 2 blanks |
| 9.5 | Test facility | glasshouse or climate room |
| 9.6 | Temperature | 24-28°C (severe test, with mild isolate)  20-24°C (mild test, with severe isolate) |
| 9.7 | Light | 12 hours per day or longer |
| 9.8 | Season | all seasons |
| 9.9 | Special measures | slightly acidic peat soil is optimal;  keep soil humid but avoid water stress |
| 10. | Inoculation |  |
| 10.1 | Preparation inoculum | aerated Messiaen or PDA or Agar Medium S of Messiaen or Czapek Dox culture or scraping of plates |
| 10.2 | Quantification inoculum | spore count, adjust to 106 spores per ml, lower concentration for a very aggressive isolate |
| 10.3 | Plant stage at inoculation | 10-18 d, cotyledon to first leaf |
| 10.4 | Inoculation method | roots and hypocotyls are immersed in spore suspension for 5‑15 min; trimming of roots is an option |
| 10.7 | Final observations | 14-21 days after inoculation |
| 11. | Observations |  |
| 11.1 | Method | visual |
| 11.2 | Observation scale | symptoms:  growth retardation, wilting, yellowing,  vessel browning extending above cotyledon |
| 11.3 | Validation of test | evaluation of variety resistance should be calibrated with results of resistant and susceptible controls. |
| 12. | Interpretation of test results in comparison with control varieties |  |
|  | absent [1] | severe symptoms |
|  | present [9] | mild or no symptoms |
| 13. | Critical control points | Test results may vary slightly in inoculum pressure due to differences in isolate, spore concentration, soil humidity and temperature. |

Proposal to change the example varieties of Characteristic 28 “Resistance to *Pyrenochaeta lycopersici* (Pl)”

*Current wording*

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
|  |  | English | français | deutsch | español | Example Varieties Exemples Beispielssorten Variedades ejemplo | Note/ Nota |
| **28.  (+)** | **VG** | **Resistance to *Pyrenochaeta lycopersici* (Pl)** | **Résistance au *Pyrenochaeta lycopersici* (Pl)** | **Resistenz gegen *Pyrenochaeta lycopersici* (Pl)** | **Resistencia a *Pyrenochaeta lycopersici* (Pl)** |  |  |
| **QL** |  | absent | absente | fehlend | Ausente | Zaralto | 1 |
|  |  | present | présente | vorhanden | Presente | Emperador | 9 |

*Proposed new wording*

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
|  |  | English | français | deutsch | español | Example Varieties Exemples Beispielssorten Variedades ejemplo | Note/ Nota |
| **28.  (+)** | **VG** | **Resistance to *Pyrenochaeta lycopersici* (Pl)** | **Résistance au *Pyrenochaeta lycopersici* (Pl)** | **Resistenz gegen *Pyrenochaeta lycopersici* (Pl)** | **Resistencia a *Pyrenochaeta lycopersici* (Pl)** |  |  |
| **QL** |  | absent | absente | fehlend | ausente | ~~Zaralto~~ | 1 |
|  |  | present | présente | vorhanden | presente | Emperador | 9 |

Proposal to change the explanation Ad. 28 in Chapter 8.2 “Explanations for individual characteristics”

*Current wording*

Ad. 28: Resistance to *Pyrenochaeta lycopersici* (Pl)

1. Pathogen *Pyrenochaeta lycopersici*

3. Host species *Solanum lycopersicum*

4. Source of inoculum -

5. Isolate -

7. Establishment pathogenicity biotest

8. Multiplication inoculum

8.1 Multiplication medium V8 Agar

8.2 Multiplication variety susceptible tomato variety

8.3 Plant stage at inoculation seed

8.4 Inoculation medium mixture of soil, e.g. (70%), sand (20%) and inoculum (10.1) (10%)

or soil mixed with diseased roots cut to small pieces

8.5 Inoculation method sowing, or transplanting at fruit maturity

8.6 Harvest of inoculum diseased roots are harvested after 2-4 months

8.7 Check of harvested inoculum visual inspection of lesions on roots

8.8 Shelf-life/viability inoculum the fungus will not die quickly, but may lose its pathogenicity

within a week after isolation on an agar medium

9. Format of the test

9.1 Number of plants per genotype 20 plants

9.2 Number of replicates……………… 1 replicate

9.3 Control varieties

susceptible: Zaralto and (*Solanum lycopersicum*) Montfavet H 63.5

resistant: Emperador and (*Solanum lycopersicum*) Kyndia, Moboglan,

Pyrella

9.5 Test facility greenhouse or climate cell

9.6 Temperature day 24°C, night 14°C

9.7 Light 12 h minimum

10. Inoculation

10.1 Preparation inoculum e.g. double-autoclaved mixture of soil with 10% oatmeal added

e.g. Incubate for 10-14 d at 20°C with occasional, repeated turning

10.3 Plant stage at inoculation 6 weeks

10.4 Inoculation method transplanting into mixture of soil, sand and inoculum (8.4)

or soil mixed with diseased roots cut to small pieces

or naturally infected soil

10.7 Final observations 6-8 weeks after transplanting (flowering plant)

11. Observations

11.1 Method visual

11.2 Observation scale Symptoms: brown lesions on roots

11.3 Validation of test evaluation of variety resistance should be calibrated with results of resistant and susceptible controls

12. Interpretation of test results in comparison with control varieties

absent ………………………… [1] symptoms

present ………………………… [9] no symptoms

13. Critical control points:

The fungus loses its pathogenicity quickly after isolation on an agar medium. It is advisable to keep the isolate alive on living plants.

*Proposed new wording*

Ad. 28: Resistance to *Pyrenochaeta lycopersici* (Pl)

|  |  |  |
| --- | --- | --- |
| 1. | Pathogen | *Pyrenochaeta lycopersici* |
| 2. | Quarantine status | No |
| 3. | Host species | *Solanum lycopersicum* |
| 4. | Source of inoculum | GEVES[[6]](#footnote-7) (FR) |
| 5. | Isolate | e.g. strain Pl 21 |
| 6. | Establishment isolate identity | On susceptible plant |
| 8. | Multiplication inoculum |  |
| 8.1 | Multiplication medium | Messiaen agar or synthetic medium |
| 8.4 | Inoculation medium | Autoclaved grains (e.g. barley) |
| 8.5 | Inoculation method | Mix of contaminated grains (e.g. 1 kg) with inoculum (e.g. medium from 2 Petri dishes with mycelium) |
| 8.6 | Harvest of inoculum | After 3 weeks |
| 9. | Format of the test |  |
| 9.1 | Number of plants per genotype | At least 20 |
| 9.2 | Number of replicates | 1 replicate |
| 9.3 | Control varieties | Susceptible : (*Solanum lycopersicum*) Marmande verte  Resistant : Emperador and (*Solanum lycopersicum*) Garance |
| 9.4 | Test design | Add non inoculated plants |
| 9.5 | Test facility | Greenhouse or climatic chamber |
| 9.6 | Temperature | 20°C |
| 9.7 | Light | At least 12h |
| 10. | Inoculation |  |
| 10.1 | Preparation inoculum | Homogenize the contaminated grains |
| 10.2 | Quantification inoculum | - |
| 10.3 | Plant stage at inoculation | 3-4 leaf stage |
| 10.4 | Inoculation method | Transplanting of plantlets in a mixture of soil (e.g. 3750 ml of soil with 750 ml of inoculum) |
| 10.7 | Final observations | 40 days post inoculation |
| 11. | Observations |  |
| 11.1 | Method | visual |
| 11.2 | Observation scale | Class 0: no necrosic lesion on roots  Class 1: few small and uncoloured necrotic lesions  Class 2: some brown necrotic lesions clearly visible (less than half the surface of the pivot)  Class 3: several brown necrotic lesions clearly visible (more than half the surface of the pivot)  Class 4: complete necrosis or destruction of the pivot |
| 11.3 | Validation of test | Evaluation of variety resistance should be calibrated with results of resistant and susceptible controls |
| 12. | Interpretation of data in terms of UPOV characteristic states | Any variety judged to be of the same resistance level or higher than Garance is judged as resistant.  Classes 0, 1 and 2 are commonly judged as resistant – Note 9  Classes 3 and 4 are commonly judged as susceptible – Note 1 |

[End of document]

1. Naktuinbouw: resistentie@naktuinbouw.nl [↑](#footnote-ref-2)
2. GEVES; Valerie.GRIMAULT@geves.fr [↑](#footnote-ref-3)
3. Naktuinbouw: resistentie@naktuinbouw.nl [↑](#footnote-ref-4)
4. GEVES: matref@geves.fr [↑](#footnote-ref-5)
5. INIA: resistencias@inia.sp [↑](#footnote-ref-6)
6. GEVES: matref@geves.fr [↑](#footnote-ref-7)