

Technical Committee

TC/55/22

Fifty-Fifth Session
Geneva, October 28 and 29, 2019Original: English
Date: October 11, 2019**PARTIAL REVISION OF THE TEST GUIDELINES FOR TOMATO***Document prepared by experts from the Netherlands**Disclaimer: this document does not represent UPOV policies or guidance*

1. The purpose of this document is to present a proposal for a partial revision of the Test Guidelines for Tomato (document TG/44/11 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 (*Solanum lycopersicum* L.) on the basis of documents TG/44/11 Rev. and TWV/53/7 "Partial revision of the Test Guidelines for Tomato" and proposed the following changes (see document TWV/53/14 Rev. "Revised Report", paragraph 93):
 - (a) To change the denomination of the races of Characteristics 48.1, 48.2 and 48.3 "Resistance to *Fusarium oxysporum* f. sp. *lycopersici* (Fol)";
 - (b) To change the explanation Ad. 48 in Chapter 8.2 "Explanations for individual characteristics";
 - (c) To change the example varieties of Characteristic 53 "Resistance to *Pyrenochaeta lycopersici* (PI)";
 - (d) To change the explanation Ad. 53 in Chapter 8.2 "Explanations for individual characteristics"
3. The proposed changes are presented from page 2 in highlight and underline (insertion) and ~~strikethrough~~ (deletion).
4. 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 should be updated in the final adopted version of this partial revision of the Test Guidelines for Tomato as follows (see document TWV/53/14 Rev. "Revised Report", paragraph 94):
 - Geves: matref@geves.fr
 - Naktuinbouw: resistentie@naktuinbouw.nl
 - INIA: resistencias@inia.sp

Proposal to change the denomination of the races of Characteristics 48.1, 48.2 and 48.3 “Resistance to *Fusarium oxysporum* f. sp. *lycopersici* (Fol)”

Current wording

	English	français	deutsch	español	Example Varieties Exemples Beispielssorten Variedades ejemplo	Note/ Nota
48. (+)	VG Resistance to <i>Fusarium oxysporum</i> f. sp. <i>lycopersici</i> (Fol)	Résistance à <i>Fusarium oxysporum</i> f. sp. <i>lycopersici</i> (Fol)	Resistenz gegen <i>Fusarium oxysporum</i> f. sp. <i>lycopersici</i> (Fol)	Resistencia a <i>Fusarium oxysporum</i> f. sp. <i>lycopersici</i> (Fol)		
48.1 (*)	VG – Race 0 (ex 1)	– Pathotype 0 (ex 1)	– Pathotyp 0 (ex 1)	– Raza 0 (ex 1)		
QL	absent	absente	fehlend	ausente	Marmande verte	1
	present	présente	vorhanden	presente	Anabel, Marporum, Marsol	9
48.2 (*)	VG – Race 1 (ex 2)	– Pathotype 1 (ex 2)	– Pathotyp 1 (ex 2)	– Raza 1 (ex 2)		
QL	absent	absente	fehlend	ausente	Marmande verte	1
	present	présente	vorhanden	presente	Motelle, Walter	9
48.3	VG – Race 2 (ex 3)	– Pathotype 2 (ex 3)	– Pathotyp 2 (ex 3)	– Raza 2 (ex 3)		
QL	absent	absente	fehlend	ausente	Marmande verte, Motelle	1
	present	présente	vorhanden	presente	Alliance, Florida, Ivanhoé, Tributes	9

Proposed new wording

	English	français	deutsch	español	Example Varieties Exemples Beispielssorten Variedades ejemplo	Note/ Nota
48. (+)	VG Resistance to <i>Fusarium oxysporum</i> f. sp. <i>lycopersici</i> (Fol)	Résistance à <i>Fusarium oxysporum</i> f. sp. <i>lycopersici</i> (Fol)	Resistenz gegen <i>Fusarium oxysporum</i> f. sp. <i>lycopersici</i> (Fol)	Resistencia a <i>Fusarium oxysporum</i> f. sp. <i>lycopersici</i> (Fol)		
48.1 (*)	VG – Race 0 (ex 1) <u>0EU/1US</u>	– Pathotype 0 (ex 1) <u>0EU/1US</u>	– Pathotyp 0 (ex 1) <u>0EU/1US</u>	– Raza 0 (ex 1) <u>0 EU/1US</u>		
QL	absent	absente	fehlend	ausente	Marmande verte	1
	present	présente	vorhanden	presente	Anabel, Marporum, Marsol	9
48.2 (*)	VG – Race 1 (ex 2) <u>1EU/2US</u>	– Pathotype 1 (ex 2) <u>1EU/2US</u>	– Pathotyp 1 (ex 2) <u>1EU/2US</u>	– Raza 1 (ex 2) <u>1EU/2US</u>		
QL	absent	absente	fehlend	ausente	Marmande verte	1
	present	présente	vorhanden	presente	Motelle, Walter	9
48.3	VG – Race 2 (ex 3) <u>2EU/3US</u>	– Pathotype 2 (ex 3) <u>2EU/3US</u>	– Pathotyp 2 (ex 3) <u>2EU/3US</u>	– Raza 2 (ex 3) <u>2EU/3US</u>		
QL	absent	absente	fehlend	ausente	Marmande verte, Motelle	1
	present	présente	vorhanden	presente	Alliance, Florida, Ivanhoé, Tributes	9

Proposal to change the explanation Ad. 48 in Chapter 8.2 “Explanations for individual characteristics”

Current wording

Ad. 48: Resistance to *Fusarium oxysporum* f. sp. *lycopersici* (Fol)

1. Pathogen *Fusarium oxysporum* f. sp. *lycopersici*
3. Host species..... *Solanum lycopersicum*
4. Source of inoculum..... Naktuinbouw¹ (NL) and GEVES² (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..... Marmande, Marmande verte, Resal
Resistant for race 0 only..... Marporum, Larissa, “Marporum x Marmande verte”, Marsol, Anabel
Resistant for race 0 and 1..... Motelle, Gourmet, Mohawk
Control varieties for the test with
race 1 (ex 2)
Susceptible..... Marmande verte, Cherry Belle, Roma
Resistant for race 0 only Marporum, Ranco
Resistant for race 0 and 1..... Tradiro, Odisea
Remark: Ranco is slightly less resistant than Tradiro
Control varieties for the test with
race 2 (ex 3)
Susceptible for race 0, 1 and 2..... Marmande verte, Motelle, Marporum
Resistant for race 0, 1 and 2..... 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 inoculums..... aerated Messiaen or PDA or Agar Medium S of Messiaen or
Czapek Dox culture or scraping of plates
- 10.2 Quantification inoculums..... pore 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. Standards near borderline R/S will help to compare between labs.
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.

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² GEVES; Valerie.GRIMAULT@geves.fr

Proposed new wording

Ad. 48: Resistance to *Fusarium oxysporum* f. sp. *lycopersici* (Fol)

1.	Pathogen	<i>Fusarium oxysporum</i> f. sp. <i>lycopersici</i>
3.	Host species	<i>Solanum lycopersicum</i>
4.	Source of inoculum	Naktuinbouw ³ (NL), GEVES ⁴ (FR) or INIA ⁵ (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 10 ⁶ 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	Marmande, Marmande verte, Resal, <u>Money maker</u>
	Resistant for race 0 only	Marporum, Larissa, "Marporum x Marmande verte", <u>Marsol</u> , <u>Anabel</u> , <u>Motelle</u> , <u>Gourmet</u> , <u>Mohawk</u> , <u>Tradiro</u>
	Resistant for race 0 and 1	<u>Motelle</u> , <u>Gourmet</u> , <u>Mohawk</u>
9.3.2	Control varieties for the test with race 1 (ex 2) 1EU/2US	
	Susceptible	Marmande verte, Cherry Belle, Roma, <u>Marporum</u> , <u>Ranco</u> , <u>Money maker</u>
	Resistant for race 0 only	<u>Marporum</u> , <u>Ranco</u>
	Resistant for race 0 and 1	Tradiro, Odisea, " <u>Motelle x Marmande verte</u> ", <u>Motelle</u>
9.3.3	Control varieties for the test with race 2 (ex 3) 2EU/3US	
	Susceptible for race 0, 1 and 2	Marmande verte, Motelle, Marporum
	Resistant for race 0, 1 and 2	Tributes, Murdoch, "Marmande verte x Florida", <u>Alliance</u> , <u>Florida</u> , <u>Ivanhoe</u>
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

³ Naktuinbouw: resistentie@naktuinbouw.nl

⁴ GEVES: matref@geves.fr

⁵ INIA: resistencias@inia.es

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 10 ⁶ 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. Standards near borderline R/S will help to compare between labs.
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 53 "Resistance to *Pyrenochaeta lycopersici* (PI)"

Current wording

53. (+)	VG	Resistance to <i>Pyrenochaeta lycopersici</i> (PI)	Résistance à <i>Pyrenochaeta lycopersici</i> (PI)	Resistenz gegen <i>Pyrenochaeta lycopersici</i> (PI)	Resistencia a <i>Pyrenochaeta lycopersici</i> (PI)		
QL		absent	absente	fehlend	ausente	Montfavet H 63.5	1
		present	présente	vorhanden	presente	Kyndia, Moboglan, Pyrella	9

Proposed new wording

53. (+)	VG	Resistance to <i>Pyrenochaeta lycopersici</i> (PI)	Résistance à <i>Pyrenochaeta lycopersici</i> (PI)	Resistenz gegen <i>Pyrenochaeta lycopersici</i> (PI)	Resistencia a <i>Pyrenochaeta lycopersici</i> (PI)		
QL		absent	absente	fehlend	ausente	<u>Montfavet H 63.5</u> <u>Marmande verte</u>	1
		present	présente	vorhanden	presente	<u>Kyndia, Moboglan, Pyrella</u> <u>Garance</u>	9

Proposal to change the explanation Ad. 53 in Chapter 8.2 "Explanations for individual characteristics"*Current wording*Ad. 53: Resistance to *Pyrenochaeta lycopersici* (PI)

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 Montfavet H 63.5
 - Resistant 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. 53: Resistance to *Pyrenochaeta lycopersici* (PI)

1.	Pathogen	<i>Pyrenochaeta lycopersici</i>
2.	Quarantine status	No
3.	Host species	<i>Solanum lycopersicum</i>
4.	Source of inoculum	GEVES ⁶ (FR)
5.	Isolate	e.g. strain PI 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: Marmande verte Resistant: Garance and (<i>S. lycopersicum</i> x <i>S. habrochaites</i>) Emperador
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
10.3	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 necrotic 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]

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