



These Test Guidelines have been superseded by a later version. The latest adopted version of Test Guidelines can be found at [http://www.upov.int/test\\_guidelines/en/list.jsp](http://www.upov.int/test_guidelines/en/list.jsp)

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Ces principes directeurs d'examen ont été remplacés par une version ultérieure. La version adoptée la plus récente des principes directeurs d'examen figure à l'adresse suivante : [http://www.upov.int/test\\_guidelines/fr/list.jsp](http://www.upov.int/test_guidelines/fr/list.jsp)

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Diese Prüfungsrichtlinien wurden durch eine neuere Fassung ersetzt. Die neueste angenommene Fassung von Prüfungsrichtlinien ist unter [http://www.upov.int/test\\_guidelines/de/list.jsp](http://www.upov.int/test_guidelines/de/list.jsp) zu finden.

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Las presentes directrices de examen han sido reemplazadas por una versión posterior. La versión de las directrices de examen de más reciente aprobación está disponible en [http://www.upov.int/test\\_guidelines/es/list.jsp](http://www.upov.int/test_guidelines/es/list.jsp).

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## INTERNATIONAL UNION FOR THE PROTECTION OF NEW VARIETIES OF PLANTS

Geneva

## TOMATO ROOTSTOCKS

UPOV Code: SOLAN\_HAB; SOLAN\_LHA;  
SOLAN\_LPE; SOLAN\_LCH; SOLAN\_PHA

*Solanum habrochaites* S. Knapp & D.M. Spooner;  
*Solanum lycopersicum* L. x *Solanum habrochaites* S.  
 Knapp & D.M. Spooner;  
*Solanum lycopersicum* L. x  
*Solanum peruvianum* (L.) Mill.;  
*Solanum lycopersicum* L. x  
*Solanum cheesmaniae* (L. Ridley) Fosberg;  
*Solanum pimpinellifolium* L. x *Solanum habrochaites*  
 S. Knapp & D.M. Spooner

## GUIDELINES

## FOR THE CONDUCT OF TESTS

## FOR DISTINCTNESS, UNIFORMITY AND STABILITY

Alternative Names:<sup>\*</sup>

Botanical name	English	French	German	Spanish
<i>Solanum habrochaites</i> S. Knapp & D.M. Spooner, <i>Lycopersicon agrimoniifolium</i> Dunal, <i>Lycopersicon hirsutum</i> Dunal, <i>Lycopersicon hirsutum</i> f. <i>glabratum</i> C. H. Müll.				
<i>Solanum lycopersicum</i> L. x <i>Solanum habrochaites</i> S. Knapp & D.M. Spooner				
<i>Solanum lycopersicum</i> L. x <i>Solanum peruvianum</i> (L.) Mill.				
<i>Solanum lycopersicum</i> L. x <i>Solanum cheesmaniae</i> (L. Ridley) Fosberg				
<i>Solanum pimpinellifolium</i> L. x <i>Solanum habrochaites</i> S. Knapp & D.M. Spooner				

The purpose of these guidelines ("Test Guidelines") is to elaborate the principles contained in the General Introduction (document TG/1/3), and its associated TGP documents, into detailed practical guidance for the harmonized examination of distinctness, uniformity and stability (DUS) and, in particular, to identify appropriate characteristics for the examination of DUS and production of harmonized variety descriptions.

## ASSOCIATED DOCUMENTS

These Test Guidelines should be read in conjunction with the General Introduction and its associated TGP documents.

Other associated UPOV documents: TG/44: Tomato (*Solanum lycopersicum* L.)

<sup>\*</sup> These names were correct at the time of the introduction of these Test Guidelines but may be revised or updated. [Readers are advised to consult the UPOV Code, which can be found on the UPOV Website (www.upov.int), for the latest information.]

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## 1. Subject of these Test Guidelines

1.1 These Test Guidelines apply to all varieties of *Solanum habrochaites* S. Knapp & D.M. Spooner; *Solanum lycopersicum* L. x *Solanum habrochaites* S. Knapp & D.M. Spooner, *Solanum lycopersicum* L. x *Solanum peruvianum* L. (Mill.), *Solanum lycopersicum* L. x *Solanum cheesmaniae* (L. Ridley) Fosberg and *Solanum pimpinellifolium* L. x *Solanum habrochaites* S. Knapp & D.M. Spooner. Such varieties are generally used as rootstocks for tomato varieties (varieties of *Solanum lycopersicum* L. (*Lycopersicum esculentum* L. (Mill.)).

1.2 Rootstocks belonging to *Solanum lycopersicum* L. (*Lycopersicum esculentum* Mill.) or to *Solanum lycopersicum* L. x *Solanum pimpinellifolium* L. (*Lycopersicum esculentum* Mill. x *Lycopersicum pimpinellifolium* Mill.) should be covered by UPOV Test Guidelines TG/44.

## 2. Material Required

2.1 The competent authorities decide on the quantity and quality of the plant material required for testing the variety and when and where it is to be delivered. Applicants submitting material from a State other than that in which the testing takes place must ensure that all customs formalities and phytosanitary requirements are complied with.

2.2 The material is to be supplied in the form of seed.

2.3 The minimum quantity of plant material, to be supplied by the applicant, should be:

10 g or 2,500 seeds.

In the case of seed, the seed should meet the minimum requirements for germination, species and analytical purity, health and moisture content, specified by the competent authority.

2.4 The plant material supplied should be visibly healthy, not lacking in vigor, nor affected by any important pest or disease.

2.5 The plant material should not have undergone any treatment which would affect the expression of the characteristics of the variety, unless the competent authorities allow or request such treatment. If it has been treated, full details of the treatment must be given.

## 3. Method of Examination

### 3.1 *Number of Growing Cycles*

The minimum duration of tests should normally be two independent growing cycles.

### 3.2 *Testing Place*

Tests are normally conducted at one place. In the case of tests conducted at more than one place, guidance is provided in TGP/9 "Examining Distinctness".

### 3.3 *Conditions for Conducting the Examination*

The tests should be carried out under conditions ensuring satisfactory growth for the expression of the relevant characteristics of the variety and for the conduct of the examination.

### 3.4 *Test Design*

3.4.1 Each test should be designed to result in a total of at least 20 plants, which should be divided between at least two replicates.

3.4.2 When resistance characteristics are used for assessing distinctness, uniformity and stability, records must be taken under conditions of controlled infection and, unless otherwise specified, on at least 20 plants.

3.4.3 The design of the tests should be such that plants or parts of plants may be removed for measurement or counting without prejudice to the observations which must be made up to the end of the growing cycle.

### 3.5 *Additional Tests*

Additional tests, for examining relevant characteristics, may be established.

## 4. Assessment of Distinctness, Uniformity and Stability

### 4.1 *Distinctness*

#### 4.1.1 General Recommendations

It is of particular importance for users of these Test Guidelines to consult the General Introduction prior to making decisions regarding distinctness. However, the following points are provided for elaboration or emphasis in these Test Guidelines.

#### 4.1.2 Consistent Differences

The differences observed between varieties may be so clear that more than one growing cycle is not necessary. In addition, in some circumstances, the influence of the environment is not such that more than a single growing cycle is required to provide assurance that the differences observed between varieties are sufficiently consistent. One means of ensuring that a difference in a characteristic, observed in a growing trial, is sufficiently consistent is to examine the characteristic in at least two independent growing cycles.

#### 4.1.3 Clear Differences

Determining whether a difference between two varieties is clear depends on many factors, and should consider, in particular, the type of expression of the characteristic being examined, i.e. whether it is expressed in a qualitative, quantitative, or pseudo-qualitative manner. Therefore, it is important that users of these Test Guidelines are familiar with the recommendations contained in the General Introduction prior to making decisions regarding distinctness.

#### 4.1.4 Number of Plants / Parts of Plants to be Examined

Unless otherwise indicated, for the purposes of distinctness, all observations on single plants should be made on 10 plants or parts taken from each of 10 plants and any other observations made on all plants in the test disregarding any off-type plants.

#### 4.1.5 Method of Observation

The recommended method of observing the characteristic for the purposes of distinctness is indicated by the following key in the second column of the Table of Characteristics (see document TGP/9 "Examining Distinctness", Section 4 "Observation of characteristics"):

- MG: single measurement of a group of plants or parts of plants
- MS: measurement of a number of individual plants or parts of plants
- VG: visual assessment by a single observation of a group of plants or parts of plants
- VS: visual assessment by observation of individual plants or parts of plants

Type of observation: visual (V) or measurement (M)

"Visual" observation (V) is an observation made on the basis of the expert's judgment. For the purposes of this document, "visual" observation refers to the sensory observations of the experts and, therefore, also includes smell, taste and touch. Visual observation includes observations where the expert uses reference points (e.g. diagrams, example varieties, side-by-side comparison) or non-linear charts (e.g. color charts). Measurement (M) is an objective observation against a calibrated, linear scale e.g. using a ruler, weighing scales, colorimeter, dates, counts, etc.

Type of record: for a group of plants (G) or for single, individual plants (S)

For the purposes of distinctness, observations may be recorded as a single record for a group of plants or parts of plants (G), or may be recorded as records for a number of single, individual plants or parts of plants (S). In most cases, “G” provides a single record per variety and it is not possible or necessary to apply statistical methods in a plant-by-plant analysis for the assessment of distinctness.

In cases where more than one method of observing the characteristic is indicated in the Table of Characteristics (e.g. VG/MG), guidance on selecting an appropriate method is provided in document TGP/9, Section 4.2.

#### 4.2 *Uniformity*

4.2.1 It is of particular importance for users of these Test Guidelines to consult the General Introduction prior to making decisions regarding uniformity. However, the following points are provided for elaboration or emphasis in these Test Guidelines:

4.2.2 For the assessment of uniformity, a population standard of 1% and an acceptance probability of at least 95% should be applied. In the case of a sample size of 20 plants, 1 off-type is allowed.

#### 4.3 *Stability*

4.3.1 In practice, it is not usual to perform tests of stability that produce results as certain as those of the testing of distinctness and uniformity. However, experience has demonstrated that, for many types of variety, when a variety has been shown to be uniform, it can also be considered to be stable.

4.3.2 Where appropriate, or in cases of doubt, stability may be further examined by testing a new seed or plant stock to ensure that it exhibits the same characteristics as those shown by the initial material supplied.

### 5. Grouping of Varieties and Organization of the Growing Trial

5.1 The selection of varieties of common knowledge to be grown in the trial with the candidate varieties and the way in which these varieties are divided into groups to facilitate the assessment of distinctness are aided by the use of grouping characteristics.

5.2 Grouping characteristics are those in which the documented states of expression, even where produced at different locations, can be used, either individually or in combination with other such characteristics: (a) to select varieties of common knowledge that can be excluded from the growing trial used for examination of distinctness; and (b) to organize the growing trial so that similar varieties are grouped together.

5.3 The following have been agreed as useful grouping characteristics:

- (a) Fruit: green shoulder (characteristic 11)
- (b) Autonecrosis (characteristic 21)
- (c) Resistance to *Meloidogyne incognita* (characteristic 22)
- (d) Resistance to *Verticillium* sp. – Race 0 (characteristic 23)
- (e) Resistance to *Fusarium oxysporum* f. sp. *lycopersici* – Race 0EU/1US (characteristic 24.1)
- (f) Resistance to *Fusarium oxysporum* f. sp. *lycopersici* – Race 1EU/2US (characteristic 24.2)
- (g) Resistance to *Fusarium oxysporum* f. sp. *lycopersici* – Race 2EU/3US (characteristic 24.3)

5.4 Guidance for the use of grouping characteristics, in the process of examining distinctness, is provided through the General Introduction and document TGP/9 “Examining Distinctness”.

## 6. Introduction to the Table of Characteristics

### 6.1 *Categories of Characteristics*

#### 6.1.1 Standard Test Guidelines Characteristics

Standard Test Guidelines characteristics are those which are approved by UPOV for examination of DUS and from which members of the Union can select those suitable for their particular circumstances.

#### 6.1.2 Asterisked Characteristics

Asterisked characteristics (denoted by \*) are those included in the Test Guidelines which are important for the international harmonization of variety descriptions and should always be examined for DUS and included in the variety description by all members of the Union, except when the state of expression of a preceding characteristic or regional environmental conditions render this inappropriate.

### 6.2 *States of Expression and Corresponding Notes*

6.2.1 States of expression are given for each characteristic to define the characteristic and to harmonize descriptions. Each state of expression is allocated a corresponding numerical note for ease of recording of data and for the production and exchange of the description.

6.2.2 In the case of qualitative and pseudo-qualitative characteristics (see Chapter 6.3), all relevant states of expression are presented in the characteristic. However, in the case of quantitative characteristics with 5 or more states, an abbreviated scale may be used to minimize the size of the Table of Characteristics. For example, in the case of a quantitative characteristic with 9 states, the presentation of states of expression in the Test Guidelines may be abbreviated as follows:

State	Note
small	3
medium	5
large	7

However, it should be noted that all of the following 9 states of expression exist to describe varieties and should be used as appropriate:

State	Note
very small	1
very small to small	2
small	3
small to medium	4
medium	5
medium to large	6
large	7
large to very large	8
very large	9

6.2.3 Further explanation of the presentation of states of expression and notes is provided in document TGP/7 "Development of Test Guidelines".

### 6.3 *Types of Expression*

An explanation of the types of expression of characteristics (qualitative, quantitative and pseudo-qualitative) is provided in the General Introduction.

### 6.4 *Example Varieties*

Where appropriate, example varieties are provided to clarify the states of expression of each characteristic.

6.5 *Legend*

- (\*) Asterisk characteristic – see Chapter 6.1.2
- QL Qualitative characteristic – see Chapter 6.3
- QN Quantitative characteristic – see Chapter 6.3
- PQ Pseudo-qualitative characteristic – see Chapter 6.3
  
- MG, MS, VG, VS – see Chapter 4.1.5
  
- (a)-(c) See Explanations on the Table of Characteristics in Chapter 8.1.
  
- (+) See Explanations on the Table of Characteristics in Chapter 8.2.



7. Table of Characteristics/Tableau des caractères/Merkmalstabelle/Tabla de caracteres

	English	français	deutsch	español	Example Varieties Exemples Beispielssorten Variedades ejemplo	Note/ Nota
<b>1. VG (*) (+)</b>	<b>Seedling: anthocyanin coloration of hypocotyl</b>	<b>Plantule: pigmentation anthocyanique de l'hypocotyle</b>	<b>Sämling: Anthocyanfärbung des Hypocotyls</b>	<b>Plántula: pigmentación antocianica del hipocótilo</b>		
<b>QL</b>	absent	absente	fehlend	ausente		1
	present	présente	vorhanden	presente	Beaufort	9
<b>2. VG (*) (+)</b>	<b>Plant: height</b>	<b>Plante: hauteur</b>	<b>Pflanze: Höhe</b>	<b>Planta: altura</b>		
<b>QN</b>	short	basse	niedrig	baja	Big Power	3
	medium	moyenne	mittel	media	Maxifort	5
	tall	haute	hoch	alta	Beaufort	7
<b>3. VG (*) (+)</b>	<b>Stem: anthocyanin coloration of upper third</b>	<b>Tige: pigmentation anthocyanique du tiers supérieur</b>	<b>Stängel: Anthocyanfärbung des oberen Drittels</b>	<b>Tallo: pigmentación antocianica del tercio superior</b>		
<b>QN (a)</b>	absent or very weak	absente ou très faible	fehlend oder sehr gering	ausente o muy débil		1
	weak	faible	gering	débil	Arnold	3
	medium	moyenne	mittel	media	Beaufort	5
	strong	forte	stark	fuerte	Montezuma	7
<b>4. VG/MS (*) (+)</b>	<b>Stem: length of internode</b>	<b>Tige: longueur de l'entre-nœud</b>	<b>Stängel: Internodienlänge</b>	<b>Tallo: longitud del entrenudo</b>		
<b>QN (a)</b>	short	court	kurz	corta	Big Force	3
	medium	moyen	mittel	media	Maxifort	5
	long	long	lang	larga	Beaufort	7
<b>5. VG/MS (*) (+)</b>	<b>Leaf: length</b>	<b>Feuille: longueur</b>	<b>Blatt: Länge</b>	<b>Hoja: longitud</b>		
<b>QN (a)</b>	short	courte	kurz	corta		3
	medium	moyenne	mittel	media	Body	5
	long	longue	lang	larga	Maxifort	7
<b>6. VG/MS (*) (+)</b>	<b>Leaf: width</b>	<b>Feuille: largeur</b>	<b>Blatt: Breite</b>	<b>Hoja: anchura</b>		
<b>QN (a)</b>	narrow	étroite	schmal	estrecha		3
	medium	moyenne	mittel	media	Body	5
	broad	large	breit	ancha	Emperador	7
<b>7. VG (*) (+)</b>	<b>Leaf: size of leaflets</b>	<b>Feuille: taille des folioles</b>	<b>Blatt: Größe der Blättfiedern</b>	<b>Hoja: tamaño de los folíolos</b>		
<b>QN (a)</b>	very small	très petites	sehr klein	muy pequeños		1
	small	petites	klein	pequeños	Titron	3
	medium	moyennes	mittel	medios	Big Force	5
	large	grandes	groß	grandes	Beaufort	7
	very large	très grandes	sehr groß	muy grandes	Hires 1210	9

	English	français	deutsch	español	Example Varieties Exemples Beispielssorten Variedades ejemplo	Note/ Nota
<b>8.</b>	<b>VG</b>	<b>Leaf: intensity of green color</b>	<b>Feuille: intensité de la couleur verte</b>	<b>Blatt: Intensität der Grünfärbung</b>	<b>Hoja: intensidad del color verde</b>	
<b>(*)</b>						
<b>QN</b>	<b>(a)</b>	light	clair	hell	claro	3
		medium	moyenne	mittel	medio	5
		dark	foncée	dunkel	oscuro	Maxifort 7
<b>9.</b>	<b>VG</b>	<b>Leaf: glossiness</b>	<b>Feuille: brillance</b>	<b>Blatt: Glanz</b>	<b>Hoja: brillo</b>	
<b>(+)</b>						
<b>QN</b>	<b>(a)</b>	weak	faible	gering	débil	Montezuma 1
		medium	moyenne	mittel	medio	Titron 2
		strong	forte	stark	fuerte	Maxifort 3
<b>10.</b>	<b>VG</b>	<b>Leaf: blistering</b>	<b>Feuille: clôture</b>	<b>Blatt: Blasigkeit</b>	<b>Hoja: abullonado</b>	
<b>(+)</b>						
<b>QN</b>	<b>(a)</b>	weak	faible	gering	débil	Montezuma 1
		medium	moyenne	mittel	medio	Emperador 2
		strong	forte	stark	fuerte	Body 3
<b>11.</b>	<b>VG</b>	<b>Fruit: green shoulder</b>	<b>Fruit : collet vert</b>	<b>Frucht: grüne Schulter</b>	<b>Fruto: hombro verde</b>	
<b>(*)</b>						
<b>QL</b>	<b>(c)</b>	absent	absent	fehlend	ausente	1
		present	présent	vorhanden	presente	Big Force, Maxifort 9
<b>12.</b>	<b>VG</b>	<b>Fruit: extent of green shoulder</b>	<b>Fruit : taille du collet vert</b>	<b>Frucht: Größe der grünen Schulter</b>	<b>Fruto: tamaño del hombro verde</b>	
<b>(*)</b>						
<b>(+)</b>						
<b>QN</b>	<b>(c)</b>	small	petit	klein	pequeño	Big Force 3
		medium	moyen	mittel	medio	5
		large	grand	groß	grande	Maxifort 7
<b>13.</b>	<b>VG</b>	<b>Fruit: intensity of green color of shoulder</b>	<b>Fruit : intensité de la couleur verte du collet</b>	<b>Frucht: Intensität der Grünfärbung der Schulter</b>	<b>Fruto: intensidad del color verde del hombro</b>	
<b>(*)</b>						
<b>QN</b>	<b>(c)</b>	light	clair	hell	claro	3
		medium	moyenne	mittel	medio	5
		dark	foncée	dunkel	oscuro	He-man 7
<b>14.</b>	<b>VG</b>	<b>Fruit: conspicuousness of meridian stripes</b>	<b>Fruit : netteté des stries médianes</b>	<b>Frucht: Ausprägung des Mittelstreifens</b>	<b>Fruto: visibilidad de las franjas meridianas</b>	
<b>(+)</b>						
<b>QN</b>	<b>(c)</b>	very weak	très faible	sehr gering	muy débil	He Wolf 1
		weak	faible	gering	débil	Popeye 2
		medium	moyenne	mittel	medio	Body 3
		strong	forte	stark	fuerte	Vigomax 4
		very strong	très forte	sehr stark	muy fuerte	5

	English	français	deutsch	español	Example Varieties Exemples Beispielssorten Variedades ejemplo	Note/ Nota	
<b>15.</b>	<b>VG/ MS</b>	<b>Pedicel: length</b>	<b>Pédicelle: longueur</b>	<b>Blütenstiel: Länge</b>	<b>Pedículo: longitud</b>		
<b>(+)</b>							
<b>QN</b>	<b>(b)</b>	short	court	kurz	corta	Titron	3
		medium	moyen	mittel	media	Multifort	5
		long	long	lang	larga	Beaufort	7
<b>16.</b>	<b>VG</b>	<b>Fruit: size</b>	<b>Fruit : taille</b>	<b>Frucht: Größe</b>	<b>Fruto: tamaño</b>		
<b>(*)</b>							
<b>(+)</b>							
<b>QN</b>	<b>(b)</b>	not developed or very small	non développé ou très petit	nicht entwickelt oder sehr klein	no desarrollado o muy pequeño	RT303	1
		small	petit	klein	pequeño	Body, Optifort	3
		medium	moyen	mittel	medio	Emperador	5
		large	grand	groß	grande	Titron	7
<b>17.</b>	<b>VG</b>	<b>Fruit: shape in longitudinal section</b>	<b>Fruit : forme en section longitudinale</b>	<b>Frucht: Form im Längsschnitt</b>	<b>Fruto: forma en sección longitudinal</b>		
<b>(*)</b>							
<b>(+)</b>							
<b>PQ</b>	<b>(b)</b>	broad oblate	aplatie large	breit breitrund	achatada ancha	He-Wolf	1
		narrow oblate	aplatie étroite	schmal breitrund	achatada estrecha	Gladiator	2
		circular	circulaire	kreisförmig	circular	Maxifort	3
		obovate	obovale	verkehrt eiförmig	obovado		4
<b>18.</b>	<b>VG/ MS</b>	<b>Fruit: number of locules</b>	<b>Fruit : nombre de loges</b>	<b>Frucht: Anzahl Kammern</b>	<b>Fruto: número de lóculos</b>		
<b>(*)</b>							
<b>QN</b>	<b>(b)</b>	only two	seulement deux	nur zwei	sólo dos	Maxifort	1
		two and three	deux et trois	zwei und drei	dos y tres		2
<b>19.</b>	<b>VG</b>	<b>Fruit: color at maturity</b>	<b>Fruit : couleur à maturité</b>	<b>Frucht: Farbe bei der Reife</b>	<b>Fruto: color en la madurez</b>		
<b>(*)</b>							
<b>PQ</b>	<b>(b)</b>	green	verte	grün	verde	Big Force	1
		yellowish	jaunâtre	gelblich	amarillento	Vigomax	2
		orangish	orangé	orangerot	anaranjado	Titron	3
		reddish	rougeâtre	rötlich	rojizo	Brigeor	4
<b>20.</b>	<b>MG</b>	<b>Time of flowering</b>	<b>Époque de floraison</b>	<b>Zeitpunkt der Blüte</b>	<b>Época de floración</b>		
<b>QN</b>		early	précoce	früh	temprana	He-Man	3
		medium	moyenne	mittel	medio	Body	5
		late	tardive	spät	tardía	Popeye	7
<b>21.</b>	<b>VG</b>	<b>Autonecrosis</b>	<b>Autonécrose</b>	<b>Autonekrose</b>	<b>Autonecrosis</b>		
<b>(*)</b>							
<b>(+)</b>							
<b>QL</b>		absent	absente	fehlend	ausente	Maxifort	1
		present	présente	vorhanden	presente	Body	9

	English	français	deutsch	español	Example Varieties Exemples Beispielsorten Variedades ejemplo	Note/ Nota
<b>22. (*)(+)</b>	<b>VG Resistance to <i>Meloidogyne incognita</i> (Mi)</b>	<b>Résistance à <i>Meloidogyne incognita</i> (Mi)</b>	<b>Resistenz gegen <i>Meloidogyne incognita</i> (Mi)</b>	<b>Resistencia a <i>Meloidogyne incognita</i> (Mi)</b>		
<b>QN</b>	susceptible	sensible	anfällig	susceptible	Bruce	1
	moderately resistant	moyennement résistant	mäßig resistent	moderadamente resistente		2
	highly resistant	hautement résistant	hoch resistent	muy resistente	Emperador	3
<b>23. (*)(+)</b>	<b>VG Resistance to <i>Verticillium</i> sp. (Va and Vd)</b>	<b>Résistance à <i>Verticillium</i> sp. (Va et Vd)</b>	<b>Resistenz gegen <i>Verticillium</i> sp. (Va und Vd)</b>	<b>Resistencia a <i>Verticillium</i> sp. (Va y Vd)</b>		
	<b>– Race 0</b>	<b>– Pathotype 0</b>	<b>– Pathotyp 0</b>	<b>– Raza 0</b>		
<b>QL</b>	absent	absente	fehlend	ausente		1
	present	présente	vorhanden	presente	Big Power	9
<b>24. (+)</b>	<b>Resistance to <i>Fusarium oxysporum</i> f. sp. <i>lycopersici</i> (Fol)</b>	<b>Résistance à <i>Fusarium oxysporum</i> f. sp. <i>lycopersici</i> (Fol)</b>	<b>Resistenz gegen <i>Fusarium oxysporum</i> f. sp. <i>lycopersici</i> (Fol)</b>	<b>Resistencia a <i>Fusarium oxysporum</i> f. sp. <i>lycopersici</i> (Fol)</b>		
<b>24.1 (*)(+)</b>	<b>VG – Race 0EU/1US</b>	<b>– Race 0EU/1US</b>	<b>– Pathotyp 0EU/1US</b>	<b>– Raza 0EU/1US</b>		
<b>QL</b>	absent	absente	fehlend	ausente		1
	present	présente	vorhanden	presente	Emperador	9
<b>24.2 (*)(+)</b>	<b>VG – Race 1EU/2US</b>	<b>– Race 1EU/2US</b>	<b>– Pathotyp 1EU/2US</b>	<b>– Raza 1EU/2US</b>		
<b>QL</b>	absent	absente	fehlend	ausente		1
	present	présente	vorhanden	presente	Emperador	9
<b>24.3 (*)(+)</b>	<b>VG – Race 2EU/3US</b>	<b>– Race 2EU/3US</b>	<b>– Pathotyp 2EU/3US</b>	<b>– Raza 2EU/3US</b>		
<b>QL</b>	absent	absente	fehlend	ausente	Emperador	1
	present	présente	vorhanden	presente	Colosus	9
<b>25. (*)(+)</b>	<b>VG Resistance to <i>Fusarium oxysporum</i> f. sp. <i>radicis-lycopersici</i> (Forl)</b>	<b>Résistance à <i>Fusarium oxysporum</i> f. sp. <i>radicis-lycopersici</i> (Forl)</b>	<b>Resistenz gegen <i>Fusarium oxysporum</i> f. sp. <i>radicis-lycopersici</i> (Forl)</b>	<b>Resistencia a <i>Fusarium oxysporum</i> f. sp. <i>radicis-lycopersici</i> (Forl)</b>		
<b>QL</b>	absent	absente	fehlend	ausente	Kemerit	1
	present	présente	vorhanden	presente	Emperador	9
<b>26. (+)</b>	<b>Resistance to <i>Fulvia fulva</i> (Ff) (ex <i>Cladosporium fulvum</i>)</b>	<b>Résistance à <i>Fulvia fulva</i> (Ff) (ex <i>Cladosporium fulvum</i>)</b>	<b>Resistenz gegen <i>Fulvia fulva</i> (Ff) (ex <i>Cladosporium fulvum</i>)</b>	<b>Resistencia a <i>Fulvia fulva</i> (Ff) (ex <i>Cladosporium fulvum</i>)</b>		
<b>26.1</b>	<b>VG – Race 0</b>	<b>– Pathotype 0</b>	<b>– Pathotyp 0</b>	<b>– Raza 0</b>		
<b>QL</b>	absent	absente	fehlend	ausente	King Kong	1
	present	présente	vorhanden	presente	Bruce	9
<b>26.2</b>	<b>VG – Group A</b>	<b>– Groupe A</b>	<b>– Gruppe A</b>	<b>– Grupo A</b>		
<b>QL</b>	absent	absente	fehlend	ausente	King Kong	1
	present	présente	vorhanden	presente	Big Power	9

	English	français	deutsch	español	Example Varieties Exemples Beispielssorten Variedades ejemplo	Note/ Nota
<b>26.3</b>	<b>VG – Group B</b>	<b>– Groupe B</b>	<b>– Gruppe B</b>	<b>– Grupo B</b>		
<b>QL</b>	absent	absente	fehlend	ausente	King Kong	1
	present	présente	vorhanden	presente	Bruce	9
<b>26.4</b>	<b>VG – Group C</b>	<b>– Groupe C</b>	<b>– Gruppe C</b>	<b>– Grupo C</b>		
<b>QL</b>	absent	absente	fehlend	ausente		1
	present	présente	vorhanden	presente	Big Power	9
<b>26.5</b>	<b>VG – Group D</b>	<b>– Groupe D</b>	<b>– Gruppe D</b>	<b>– Grupo D</b>		
<b>QL</b>	absent	absente	fehlend	ausente	King Kong	1
	present	présente	vorhanden	presente	Bruce	9
<b>26.6</b>	<b>VG – Group E</b>	<b>– Groupe E</b>	<b>– Gruppe E</b>	<b>– Grupo E</b>		
<b>QL</b>	absent	absente	fehlend	ausente	Bruce, King Kong	1
	present	présente	vorhanden	presente	Big Power	9
<b>27.</b>	<b>Resistance to Tomato mosaic virus (ToMV)</b>	<b>Résistance au virus de la mosaïque de la tomate (ToMV)</b>	<b>Resistenz gegen das Tomatenmosaikvirus (ToMV)</b>	<b>Resistencia al virus del mosaico del tomate (ToMV)</b>		
<b>(+)</b>						
<b>27.1</b>	<b>VG – Strain 0</b>	<b>– Souche 0</b>	<b>– Pathotyp 0</b>	<b>– Cepa 0</b>		
<b>QL</b>	absent	absente	fehlend	ausente		1
	present	présente	vorhanden	presente	Emperador	9
<b>27.2</b>	<b>VG – Strain 1</b>	<b>– Souche 1</b>	<b>– Pathotyp 1</b>	<b>– Cepa 1</b>		
<b>QL</b>	absent	absente	fehlend	ausente		1
	present	présente	vorhanden	presente	Emperador	9
<b>27.3</b>	<b>VG – Strain 2</b>	<b>– Souche 2</b>	<b>– Pathotyp 2</b>	<b>– Cepa 2</b>		
<b>QL</b>	absent	absente	fehlend	ausente		1
	present	présente	vorhanden	presente	Emperador	9
<b>28.</b>	<b>VG Resistance to <i>Pyrenochaeta lycopersici</i> (PI)</b>	<b>Résistance au <i>Pyrenochaeta lycopersici</i> (PI)</b>	<b>Resistenz gegen <i>Pyrenochaeta lycopersici</i> (PI)</b>	<b>Resistencia a <i>Pyrenochaeta lycopersici</i> (PI)</b>		
<b>(+)</b>						
<b>QL</b>	absent	absente	fehlend	ausente		1
	present	présente	vorhanden	presente	Emperador	9
<b>29.</b>	<b>VG Resistance to <i>Stemphylium</i> spp. (Ss)</b>	<b>Résistance à <i>Stemphylium</i> spp. (Ss)</b>	<b>Resistenz gegen <i>Stemphylium</i> spp. (Ss)</b>	<b>Resistencia a <i>Stemphylium</i> spp. (Ss)</b>		
<b>(+)</b>						
<b>QL</b>	absent	absente	fehlend	ausente	Big Power	1
	present	présente	vorhanden	presente	Body	9
<b>30.</b>	<b>VG Resistance to Tomato yellow leaf curl virus (TYLCV)</b>	<b>Résistance au virus des feuilles jaunes en cuillère de la tomate (TYLCV)</b>	<b>Resistenz gegen gelbes Tomatenblattrollvirus (TYLCV)</b>	<b>Resistencia al virus del enrollamiento de la hoja (TYLCV)</b>		
<b>(+)</b>						
<b>QL</b>	absent	absente	fehlend	ausente	Big Power	1
	present	présente	vorhanden	presente		9

	English	français	deutsch	español	Example Varieties Exemples Beispielssorten Variedades ejemplo	Note/ Nota
<b>31.</b>	<b>VG</b>	<b>Resistance to Tomato spotted wilt virus (TSWV)</b>	<b>Résistance au virus de la tache bronzée de la tomate (TSWV)</b>	<b>Resistenz gegen das gefleckte Tomaten-bronzenfleckenvirus (TSWV)</b>	<b>Resistencia al virus del bronceado de tomate (TSWV)</b>	
<b>(+)</b>						
<b>QL</b>	absent	absente	fehlend	ausente	Emperador	1
	present	présente	vorhanden	presente	Enpower	9
<b>32.</b>	<b>VG</b>	<b>Resistance to <i>Oidium neolycopersici</i> (On)</b>	<b>Résistance à <i>Oidium neolycopersici</i> (On)</b>	<b>Resistenz gegen <i>Oidium neolycopersici</i> (On)</b>	<b>Resistencia a <i>Oidium neolycopersici</i> (On)</b>	
<b>(+)</b>						
<b>QL</b>	absent	absente	fehlend	ausente		1
	present	présente	vorhanden	presente	Multifort	9

## 8. Explanations on the Table of Characteristics

### 8.1 *Explanations covering several characteristics*

Characteristics containing the following key in the second column of the Table of Characteristics should be examined as indicated below:

- (a) Observations on the plant, stem and leaves should be done after a fruit set on at least five trusses and before ripening of the second truss. Observations should be done before deterioration of the leaves.
- (b) Observations on the fruit should be made on mature fruits from the second or higher truss.
- (c) Observations on the green shoulder and meridian stripes of the fruit should be made on the plant before maturity.

### 8.2 *Explanations for individual characteristics*

#### Ad. 1: Seedling: anthocyanin coloration of hypocotyl



#### Ad. 2: Plant: height

To be observed after fruit set on 5 nodes.

#### Ad. 4: Stem: length of internode

The mean length of the internodes between the 1st and 4th trusses should be assessed.

#### Ad. 7: Leaf: size of leaflets

The size of the leaflet should be observed in the middle of the leaf.

#### Ad. 9: Leaf: glossiness

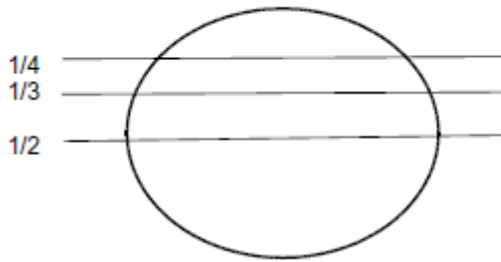
The glossiness of the leaf should be observed in the middle of the plant.

Ad. 10: Leaf: blistering

Caution is required for confusion between blistering and creasing. Blistering is the difference in height of the surface of the leaf between the veins. Creasing is independent from the veins. The blistering should be observed in the middle third of the plant.

Ad. 12: Fruit: extent of green shoulder

The gene for green shoulder might not be clearly expressed in some conditions.



3: small (1/4)  
5: medium (1/3)  
7: large (1/2)

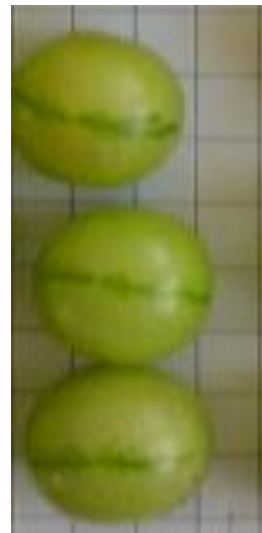
Ad. 14: Fruit: conspicuousness of meridian stripes



2  
weak



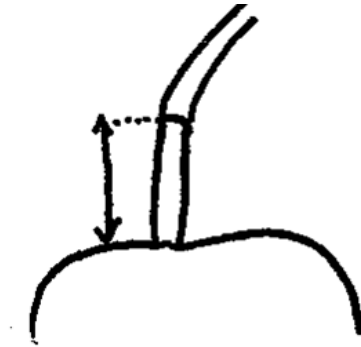
3  
medium



4  
strong



Ad. 15: Pedicel: length

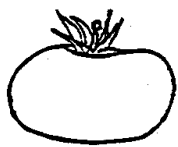


Ad. 16: Fruit: size

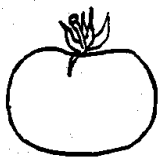
Varieties of certain interspecific crosses for tomato rootstocks, may not produce fruits, or exceptionally produce few very small fruits (note 1).

Ad. 17: Fruit: shape in longitudinal section

The apex is considered to be the part that is furthest from the stalk attachment.



1  
broad oblate



2  
narrow oblate



3  
circular



4  
obovate

Ad. 21: Autonecrosis

Autonecrosis is a necrotic reaction to the presence of incompatible genomes causing older leaves to wither and die.

Ad. 22: Resistance to *Meloidogyne incognita* (Mi)

- |   |  |
|---|--|
| 1. Pathogen .....   | <i>Meloidogyne incognita</i>   |
| 3. Host species .....   | <i>Solanum lycopersicum</i>  |
| 4. Source of inoculum .....   | Naktuinbouw (NL <sup>1</sup> ) or GEVES <sup>2</sup> (FR)  |
| 5. Isolate .....  | non-resistance breaking  |
| 6. Establishment isolate identity .....   | use rootstock or tomato standards  |
| 7. Establishment pathogenicity .....  | use susceptible rootstock or tomato standard   |
| 8. Multiplication inoculum  |  |
| 8.1 Multiplication medium .....   | living plant   |
| 8.2 Multiplication variety .....  | preferably resistant to powdery mildew   |
| 8.3 Plant stage at inoculation .....  | see 10.3   |
| 8.5 Inoculation method .....  | see 10.4   |
| 8.6 Harvest of inoculum .....   | root systems are cut with scissors into pieces<br>of about 1 cm length   |
| 8.7 Check of harvested inoculum .....   | visual check for presence of root knots  |
| 8.8 Shelf life/viability inoculum.....  | 1 day  |
| 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: .....  | Bruce and ( <i>Solanum lycopersicum</i> ) Clairvil, Casaque Rouge  |
| Moderately resistant : .....  | ( <i>Solanum lycopersicum</i> ) Madyta, Campeon, Madyta, Vinchy  |
| Highly resistant: .....   | Emperador and ( <i>Solanum lycopersicum</i> ) "Anahu x Casaque Rouge",<br>Anahu, Anabel                                  |
| 9.4 Test design .....   | include standard varieties   |
| 9.5 Test facility .....   | greenhouse or climate room   |
| 9.6 Temperature .....   | not over 28° C   |
| 9.7 Light .....   | at least 12 h per day  |
| 10. Inoculation   |  |
| 10.1 Preparation inoculum.....  | small pieces of diseased root mixed with soil<br>mix soil and infested root pieces                                       |
| 10.2 Quantification inoculum .....  | soil: root ratio = 8:1, or depending on experience   |
| 10.3 Plant stage at inoculation .....   | seed, or cotyledons  |
| 10.4 Inoculation method .....   | plants are sown in infested soil or contamination of soil after sowing<br>when plantlets are at cotyledon stage          |
| 10.7 Final observations .....   | 28 to 45 days after inoculation  |
| 11. Observations  |  |
| 11.1 Method.....  | root inspection  |
| 11.2 Observation scale .....  | Symptoms:<br>Galling, root malformation,<br>growth reduction, plant death  |
| 11.3 Validation of test .....   | evaluation of variety resistance should be calibrated with results of<br>resistant and susceptible controls on standards |
| 12. Interpretation of test results in comparison with control varieties                                       |  |
| To consider that resistant varieties may have a few plants with falls. These are not considered as off-types. |  |
| absent (susceptible).....   | [1] growth strongly reduced, high gall count   |
| intermediate<br>(moderately resistant).....   | [2] medium growth reduction, medium gall count   |
| present (highly resistant).....   | [3] no growth reduction, no galls  |
| 13. Critical control points:  |  |
| Avoid rotting of roots; high temperature causes breakdown of resistance                                       |  |

<sup>1</sup> Naktuinbouw: [resistentie@naktuinbouw.nl](mailto:resistentie@naktuinbouw.nl)

<sup>2</sup> Geves: [matref@geves.fr](mailto:matref@geves.fr)

Ad. 23: Resistance to *Verticillium* sp. (Va and Vd)

1. Pathogen ..... *Verticillium dahliae* or *Verticillium albo-atrum* (see note below)
3. Host species ..... *Solanum lycopersicum*
4. Source of inoculum ..... Naktuinbouw<sup>3</sup> (NL) and GEVES<sup>4</sup> (FR)
5. Isolate ..... Race 0 (e.g. strain Toreilles 4-1-4-1)
8. Multiplication inoculum
- 8.1 Multiplication medium ..... Potato Dextrose Agar, Agar Medium "S" of Messiaen
- 8.4 Inoculation medium..... water (for scraping agar plates) or Czapek Dox broth (3-7 d-old aerated culture at 20-25°C, in darkness)
- 8.6 Harvest of inoculum ..... filter through double muslin cloth
- 8.7 Check of harvested inoculums..... spore count; adjust to 10<sup>6</sup> per ml
- 8.8 Shelf life/viability inoculums..... 1 d at 4°C
9. Format of the test
- 9.1 Number of plants per genotype..... 35 seeds for 24 plants
- 9.2 Number of replicates..... 1 replicate
- 9.3 Control varieties
- Susceptible ..... (*Solanum lycopersicum*) Flix, Marmande verte, Clarion, Santonio, Anabel
- Resistant ..... Big Power and (*Solanum lycopersicum*) Monalbo, Elias, Monalbo x Marmande verte, Daniela, Marmande VR
- 9.4 Test design..... 20 plants inoculated at least, 2 blanks at least
- 9.5 Test facility..... greenhouse or climate room
- 9.6 Temperature..... optimal 20-25°C, 20-22°C after inoculation
- 9.7 Light..... 12 h or longer
10. Inoculation
- 10.1 Preparation inoculums..... aerated, liquid culture (8.4)
- 10.2 Quantification inoculums..... count spores, adjust to 10<sup>6</sup> per ml
- 10.3 Plant stage at inoculation..... cotyledon to 3rd leaf
- 10.4 Inoculation method..... roots are immersed for 4 to 15 min in spore suspension.
- 10.7 Final observations..... 14-33 d after inoculation
11. Observations
- 11.1 Method..... visual
- 11.2 Observation scale..... growth retardation, wilting, chlorosis, and vessel browning
- 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 laboratories.
12. Interpretation of test results in comparison with control varieties
- |         |           |                     |
|---------|-----------|---------------------|
| absent  | ..... [1] | severe symptoms     |
| present | ..... [9] | mild or no symptoms |
13. Critical control points:  
All symptoms may be present in resistant varieties, but the severity will be distinctly less than in susceptible varieties. Usually resistant varieties will show significantly less growth retardation than susceptible varieties.

<sup>3</sup> Naktuinbouw: resistantie@naktuinbouw.nl

<sup>4</sup> Geves: matref@geves.fr

Ad. 24: 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 <sup>5</sup> (NL), GEVES <sup>6</sup> (FR) or INIA <sup>7</sup> (ES)
5.	Isolate	race 0EU/1US (e.g. strains Orange 71 or PRI 20698 or Fol 071) race 1EU/2US (e.g. strains 4152 or PRI40698 or RAF 70) race 2EU/3US (e.g. strain Fol029)
6.	Establishment isolate identity	use differential varieties (see ISF website: <a href="http://www.worldseed.org">http://www.worldseed.org</a> )
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 <sup>6</sup> 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 0EU/1US	
	Susceptible	( <i>Solanum lycopersicum</i> ) Marmande, Marmande verte, Resal
	Resistant	Emperador, Colosus and ( <i>Solanum lycopersicum</i> ) "Marporum x Marmande verte", Motelle, Gourmet, Mohawk, Ranco, Tradiro
9.3.2	Control varieties for the test with race 1EU/2US	
	Susceptible	( <i>Solanum lycopersicum</i> ) Marmande verte, Cherry Belle, Roma, Marporum, Ranco
	Resistant	Emperador, Colosus and ( <i>Solanum lycopersicum</i> ) Tradiro, Odisea, "Motelle x Marmande verte"
9.3.3	Control varieties for the test with race 2EU/3US	
	Susceptible	Emperador and ( <i>Solanum lycopersicum</i> ) Marmande verte, Motelle, Marporum
	Resistant	Colosus and ( <i>Solanum lycopersicum</i> ) 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 10 <sup>6</sup> spores per ml, lower concentration for a very aggressive isolate
10.3	Plant stage at inoculation	10-18 d, cotyledon to first leaf

<sup>5</sup> Naktuinbouw: resistentie@naktuinbouw.nl

<sup>6</sup> GEVES: matref@geves.fr

<sup>7</sup> INIA: resistencias@inia.es

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.

Ad. 25: Resistance to *Fusarium oxysporum* f. sp. *radicis-lycopersici* (Forl)

1. Pathogen .....	<i>Fusarium oxysporum</i> f. sp. <i>radicis-lycopersici</i>
3. Host species .....	<i>Solanum lycopersicum</i>
4. Source of inoculum .....	Naktuinbouw <sup>8</sup> (NL) and GEVES <sup>9</sup> (FR)
5. Isolate .....	-
7. Establishment pathogenicity .....	symptoms on susceptible tomato
8. Multiplication inoculum	
8.1 Multiplication medium .....	Potato Dextrose Agar or Medium agar "S" of Messiaen
8.4 Inoculation medium.....	water for scraping agar plates or Czapek-Dox (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 <sup>6</sup> 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	
Susceptible: .....	Kemerit and ( <i>Solanum lycopersicum</i> ) Motelle, Moneymaker
Resistant: .....	Emperador and ( <i>Solanum lycopersicum</i> ) Momor, "Momor x Motelle"
Remark: .....	"Momor x Motelle" has slightly weaker resistance than Momor
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) 17-24°C (mild test, with severe isolate)
9.7 Light .....	at least 12 hours per day
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 culture or scraping of plates
10.2 Quantification inoculum .....	spore count, adjust to 10 <sup>6</sup> spores per ml
10.3 Plant stage at inoculation .....	12-18 d, cotyledon to third leaf
10.4 Inoculation method .....	roots and hypocotyls are immersed in spore suspension for 5-15 min
10.7 Final observations.....	10-21 days after inoculation
11. Observations	
11.1 Method.....	visual; a few plants are lifted at the end of the test
11.2 Observation scale .....	Symptoms: Plant death, Growth retardation caused by root degradation Root degradation, Necrotic pinpoint and necrotic lesions on stems
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:	
Temperature should never exceed 27°C during the test period; frequent renewal of races may be needed because of loss of pathogenicity	

<sup>8</sup> Naktuinbouw: resistantie@naktuinbouw.nl  
<sup>9</sup> Geves: matref@geves.fr

Ad. 26: Resistance to *Fulvia fulva* (Ff) (ex *Cladosporium fulvum*)

1. Pathogen ..... *Fulvia fulva* (ex *Cladosporium fulvum*)
  3. Host species ..... *Solanum lycopersicum*
  4. Source of inoculum ..... Naktuinbouw<sup>10</sup> (NL) or GEVES<sup>11</sup> (FR)
  5. Isolate ..... Race group 0, A, B, C, D, and E
  6. Establishment isolate identity ..... with genetically defined differentials from GEVES (FR)  
A breaks Cf-2, B Cf-4, C Cf-2&4, D Cf-5, E Cf-2&4&5
  7. Establishment pathogenicity ..... symptoms on susceptible tomato
  8. Multiplication inoculum.....
  - 8.1 Multiplication medium ..... Potato Dextrose Agar or Malt Agar or a synthetic medium
  - 8.8 Shelf life/viability inoculum..... 4 hours, keep cool
  9. Format of the test
  - 9.1 Number of plants per genotype ..... more than 20 plants
  - 9.2 Number of replicates..... 1 replicate
  - 9.3 Control varieties
  - Susceptible: ..... King Kong and (*Solanum lycopersicum*) Monalbo, Moneymaker
  - Resistant for race 0:..... Bruce and (*Solanum lycopersicum*) Angela, Estrella, Sonatine, Sonato, Vemone, Vagabond, IVT 1149, Vagabond x IVT 1149, IVT 1154
  - Resistant for race group A: ..... Big Power and (*Solanum lycopersicum*) Angela, Estrella, Sonatine, Sonato
  - Resistant for race group B: ..... Bruce and (*Solanum lycopersicum*) Angela, Estrella, Sonatine, Sonato, Vemone
  - Resistant for race group C: ..... Big Power and (*Solanum lycopersicum*) Angela, Estrella, Sonatine
  - Resistant for race group D: ..... Bruce and (*Solanum lycopersicum*) Estrella, Sonatine, Vemone
  - Resistant for race group E: ..... Big Power and (*Solanum lycopersicum*) Sonatine, Jadviga, Rhianna, IVT 1154
  - 9.5 Test facility ..... glasshouse or climate room
  - 9.6 Temperature ..... day: 22° C, night: 20° or day: 25°C, night 20°C
  - 9.7 Light ..... 12 hours or longer
  - 9.9 Special measures ..... depending on facility and weather, there may be a need to raise the humidity  
e.g. humidity tent closed 3-4 days after inoculation  
and after this, 66% until 80% closed during day, until end
  10. Inoculation .....
  - 10.1 Preparation inoculum..... prepare evenly colonized plates, e.g. 1 for 36 plants;  
remove spores from plate by scraping with water with Tween20;  
filter through double muslin cloth
  - 10.2 Quantification inoculum ..... count spores; adjust to 10<sup>5</sup> spores per ml or more
  - 10.3 Plant stage at inoculation ..... 19-20 d (incl. 12 d at 24°), 2-3 leaves
  - 10.4 Inoculation method ..... spray on dry leaves
  - 10.7 Final observations..... 14 days after inoculation
  11. Observations
  - 11.1 Method..... visual inspection of abaxial side of inoculated leaves
  - 11.2 Observation scale ..... Symptom: velvety, white spots
  - 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
- Excessively high humidity may cause rugged brown spots on all leaves. These are not to be considered as off-types.
13. Critical control points:
- Ff spores have a variable size and morphology. Small spores are also viable.  
Fungal plates will gradually become sterile after 6-10 weeks. Store good culture at -80°C.  
For practical purposes, it is not possible to keep plants longer than 14 days inside a tent.

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<sup>11</sup> Geves: matref@geves.fr

**Ad. 27: Resistance to Tomato mosaic virus (ToMV)**

Resistance to be tested in a bio-assay (method i) or in a DNA marker test (method ii), if appropriate.

**(i) Bio-assay**

1.	Pathogen	Tomato mosaic virus
3.	Host species	<i>Solanum lycopersicum</i>
4.	Source of inoculum	Naktuinbouw <sup>12</sup> (NL) or GEVES <sup>13</sup> (FR)
5.	Isolate	Strain 0 (e.g. isolate INRA Avignon 6-5-1-1), strain 1 and strain 2
6.	Establishment isolate identity	genetically defined tomato standards Mobaci (Tm1), Moperou (Tm2), Momor (Tm2 <sup>2</sup> )
7.	Establishment pathogenicity	on susceptible plant
8.	Multiplication inoculum	
8.1	Multiplication medium	living plant
8.2	Multiplication variety	e.g. Moneymaker, Marmande
8.7	Check of harvested inoculum	option: on <i>Nicotiana tabacum</i> "Xanthi", check lesions after 2 days
8.8	Shelflife/viability inoculum	fresh>1 day, desiccated>1year
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	
	Susceptible	( <i>Solanum lycopersicum</i> ) Marmande, Monalbo
	Resistant for ToMV: 0 and 2	( <i>Solanum lycopersicum</i> ) Mobaci
	Resistant for ToMV: 0 and 1	( <i>Solanum lycopersicum</i> ) Moperou
	Resistant with necrosis	( <i>Solanum lycopersicum</i> ) "Monalbo x Momor"
	Resistant	( <i>Solanum lycopersicum</i> ) Gourmet
9.4	Test design	blank treatment with PBS and carborundum or similar buffer
9.5	Test facility	glasshouse or climate room
9.6	Temperature	24 to 26°C
9.7	Light	12 hours or longer
9.8	Season	symptoms are more pronounced in summer
10.	Inoculation	
10.1	Preparation inoculum	1 g leaf with symptoms with 10 ml PBS or similar buffer homogenize, add carborundum to buffer (1 g/30ml)
10.3	Plant stage at inoculation	cotyledons or 2 leaves
10.4	Inoculation method	gentle rubbing
10.7	Final observations	11-21 days after inoculation
11.	Observations	
11.1	Method	visual
11.2	Observation scale	symptoms of susceptibility: mosaic in top, leaf malformation symptoms of resistance (based on hypersensitivity): local necrosis, top necrosis, systemic necrosis
11.3	Validation of test	evaluation of variety resistance should be calibrated with results of resistant and susceptible controls
	Remark: in some heterozygous varieties a variable proportion of plants may have severe systemic necrosis or some necrotic spots while the other plants have no symptoms. This proportion may vary between experiments.	
12.	Interpretation of test results in comparison with control varieties	
	absent .....[1]	symptoms of susceptibility
	present ..... [9]	no symptoms, or symptoms of hypersensitive resistance
13.	Critical control points	Temperature and light may influence the development of necrosis. More light means more necrosis. At temperatures above 26°C the resistance may break down. Resistant heterozygous varieties may have symptomless plants and plants with severe necrosis; in spite of apparent segregation the sample may be evaluated as uniform for resistance. Note: Strain INRA Avignon 6-5-1-1 is recommended for ToMV: 0. This strain causes a striking yellow Aucuba mosaic.

<sup>12</sup> Naktuinbouw: resistentie@naktuinbouw.nl

<sup>13</sup> GEVES: matref@geves.fr



(ii) DNA marker test

Resistance gene Tm2 gives resistance to ToMV. Gene Tm2 has two dominant alleles for resistance: allele Tm2 is always associated with resistance to strain 0 and 1, allele Tm2<sup>2</sup> is always associated with resistance to strain 0, 1 and 2. The presence or absence of both alleles for resistance can be detected by the co-dominant markers as described in Arens, P. et al (2010). Specific aspects:

1.	Pathogen	Tomato mosaic virus
2.	Functional gene	Tm2/2 <sup>2</sup>
3.	Primers	
3.1	Assay 1 to check resistance allele Tm2 or Tm2 <sup>2</sup>	Outer primer TMV-2286F: 5'GGGTATACTGGGAGTGTCCAATTC3' Outer primer TMV-2658R: 5'CCGTGCACGTTACTTCAGACAA3' Tm2 <sup>2</sup> SNP2494F: 5'CTCATCAAGCTTACTCTAGCCTACTTTAGT3' Tm2 SNP2493R: 5'CTGCCAGTATATAACGGTCTACCG3'
3.2	Assay 2 to check allele for susceptibility or resistance	Outer primer TM2-748F: 5'CGGTCTGGGGAAAACAACCTCT3' Outer primer TM2-1256R: 5'CTAGCGGTATACCTCCACATCTCC3' TM2-SNP901misR: 5'GCAGGTTGTCCTCCAAATTTTCCATC3' TM2-SNP901misF: 5'CAAATTGGACTGACGGAACAGAAAGTT3'
4.	Format of the test	
4.1	Number of plants per genotype	at least 20 plants
4.2	Control varieties	homozygous allele for susceptibility tm2 present: ( <i>Solanum lycopersicum</i> ) Moneymaker homozygous allele for resistance Tm2 present: ( <i>Solanum lycopersicum</i> ) Moperou homozygous allele for resistance Tm2 <sup>2</sup> present: Emperador
6.	PCR conditions	1. Initial denaturation step at 94°C for 3 minutes 2. 35 cycles at 94°C for 1 minute, 55°C for 1 minute, 72°C for 2 minutes 3. Final extension step of 72°C for 10 minutes
8.	Interpretation of test results	The presence of the alleles tm2, Tm2, Tm2 <sup>2</sup> lead to different interpretation for characteristics 27.1, 27.2 and 27.3, see table. In case the DNA marker test result does not confirm the declaration in the TQ, a bio-assay should be performed to observe whether the variety is resistant due to another mechanism like gene Tm1.

Test result DNA marker test	tm2/tm2	Tm2/tm2 or Tm2/Tm2	Tm2 <sup>2</sup> /tm2 or Tm2 <sup>2</sup> /Tm2 <sup>2</sup> or Tm2 <sup>2</sup> /Tm2
27.1 Strain 0	[1] absent	[9] resistant	[9] resistant
27.2 Strain 1	[1] absent	[9] resistant	[9] resistant
27.3 Strain 2	[1] absent	[1] absent	[9] resistant

**Ad. 28: 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 <sup>14</sup> (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 : ( <i>Solanum lycopersicum</i> ) Marmande verte Resistant : Emperador and ( <i>Solanum lycopersicum</i> ) 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 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

<sup>14</sup> GEVES: matref@geves.fr

Ad. 29: Resistance to *Stemphylium* spp. (Ss)

- |   |   |
|---|---|
| 1. Pathogen .....   | <i>Stemphylium</i> spp. e.g. <i>Stemphylium solani</i> (see note below)   |
| 3. Host species .....   | <i>Solanum lycopersicum</i>   |
| 4. Source of inoculum .....   | GEVES <sup>15</sup> (FR)  |
| 5. Isolate .....  | -   |
| 7. Establishment pathogenicity .....                                    | biotest   |
| 8. Multiplication inoculum  |   |
| 8.1 Multiplication medium .....   | PDA (12 hours per day under near-ultraviolet light to induce sporulation) or V8   |
| 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 .....   |   |
| Susceptible: .....  | Big Power and ( <i>Solanum lycopersicum</i> ) Monalbo   |
| Resistant: .....  | Body and ( <i>Solanum lycopersicum</i> ) Motelle, F1 Motelle x Monalbo  |
| 9.5 Test facility .....   | greenhouse or climate cell  |
| 9.6 Temperature .....   | 24°C  |
| 9.7 Light .....   | 12 hours minimum  |
| 9.9 Special measures .....  | incubation in tunnel with 100 % relative humidity or humidity tent closed 5 days after inoculation, after this, 80% until end   |
| 10. Inoculation   |   |
| 10.1 Preparation inoculum .....   | sporulating plates (8.1) are scraped and air-dried overnight<br>The next day plates are soaked and stirred for 30 min in a beaker with demineralized water, or sporulating plates are scraped with water with Tween<br>The spore suspension is sieved through a double layer of muslin. |
| 10.2 Quantification inoculum .....                                      | 5.10 <sup>3</sup> – 10 <sup>5</sup> spores per ml   |
| 10.3 Plant stage at inoculation .....                                   | 20-22 days (three expanded leaves)  |
| 10.4 Inoculation method .....   | spraying  |
| 10.7 Final observations .....   | 4 -10 days after inoculation  |
| 11. Observations  |   |
| 11.1 Method .....   | visual  |
| 11.2 Observation scale .....  | Symptoms:<br>necrotic lesions on cotyledons and leaves;<br>yellowing of leaves  |
| 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 (11.2)   |
| present .....   | [9] no symptoms, or less than resistant standard  |
| 13. Critical control points: .....                                      | 8.1 and 10.1  |

Note: Some isolates of *Stemphylium* cannot be classified easily as either *Stemphylium solani* or a related species. These *Stemphylium* isolates may still be useful for identifying resistance to *Stemphylium solani*.

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Ad. 30: Resistance to Tomato yellow leaf curl virus (TYLCV)

(i) agroinoculation method

1.	Pathogen	Tomato yellow leaf curl virus (TYLCV) IL strain. (See note below)
2.	Quarantine status	yes (see 13.)
3.	Host species	<i>Solanum lycopersicum</i>
4.	Source of inoculum	Dr. Eduardo R. Bejarano, Plant Genetics Laboratory, IHSM-UMA-CSIC <sup>16</sup>
5.	Isolate	Alm:Pep:99, strain IL
6.	Establishment isolate identity	
7.	Establishment pathogenicity	
8.	Multiplication inoculum	
8.1	Multiplication medium	YEP/Kanamycin.
8.2	Multiplication variety	
8.3	Plant stage at inoculation	3-4 leaf
8.4	Inoculation medium	YEP
8.5	Inoculation method	Stem puncture agroinfiltration. Plant agroinoculation is carried out using <i>Agrobacterium tumefaciens</i> transformed with plasmids containing the infectious clones (Morilla, et al. 2005. <i>Phytopathology</i> 95: 1089-1097) <sup>17</sup>
8.6	Harvest of inoculums	
8.7	Check of harvested inoculum	
8.8	Shelflife/viability inoculum	<i>A. tumefaciens</i> stocks are maintained frozen at -80°C in 15-20% glycerol for long term storage. Cultures to be stored are typically started from a single colony and grown in 5 ml YEP +2.5 µl kanamycin (100mg/ml) during 48 h at 28°C.
9.	Format of the test	
9.1	Number of plants per genotype	20
9.2	Number of replicates	2
9.3	Control varieties	Susceptible: Big Power, ( <i>Solanum lycopersicum</i> ) Moneymaker, Marmande Resistant: ( <i>Solanum lycopersicum</i> ) Delyca, Montenegro, Anastasia, TY20, Mohawk
9.4	Test design	
9.5	Test facility	Glasshouse or climatic chamber with permission to confined use of LMO/GMO, confinement level 1 (N-1) <sup>17</sup>
9.6	Temperature	23-25°C
9.7	Light	16 h
9.8	Season	
9.9	Special measures	Permission to confined use of LMO/GMO, at least level 1 (N-1) <sup>17</sup>
10.	Inoculation	
10.1	Preparation inoculum	Streak the surface of the frozen <i>A. tumefaciens</i> stock tube and submerge in 5 ml YEP+2.5 µl kanamycin (100mg/ml) during 48 h at 28°C. Shaking is needed. Take 100 µl and place them into 100 ml YEP and 50 µl kanamycin (100mg/ml). Shake 48 h at 28°C. Centrifuge the saturated culture for 20 min at 3500 rpm and discard supernatant.
10.2	Quantification inoculums	Dissolve in sterile deionize water to a final OD <sub>600</sub> of 1.
10.3	Plant stage at inoculation	3-4 <sup>th</sup> leaf
10.4	Inoculation method	Take up into a 1 ml syringe with a 27-gauge needle and few drops (about 20 µl of the culture) were deposited on 10-15 puncture wounds made with the needle into the stem of test tomato plants. Maintain on ice while inoculating plants.
10.5	First observation	20 days post inoculation
10.6	Second observation	30 dpi

<sup>16</sup> Source of inoculum: IHSM-UMA-CSIC, edu\_rodri@uma.es or INIA, resistencias@inia.es

<sup>17</sup> The transformed *Agrobacterium tumefaciens* is a living modified organism (LMO; or genetically modified organism (GMO)) and in many countries it requires to comply with Cartagena Protocol on Biosafety in case of transboundary movement, transit, handling and use that may have adverse effects on the conservation and sustainable use of biological diversity, taking also into account risks to human health.

*10.7	Final observations	45 dpi
11.	Observations	
11.1	Method	Visual
11.2	Observation scale	Symptoms: leaf yellowing and curling
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	
	absent.....[1]	severe symptoms
	present..... [9]	no symptoms
13.	Critical control points: TYLCV is endemic in many tropical and subtropical areas and has a quarantine status in many countries with a temperate climate. TYLCV-IL is the strain most widely spread worldwide. With this strain, symptoms do not appear in varieties with Ty-1 and Ty-2. TYLCV is on the EPPO alert list. Some TYLCV resistant varieties may be susceptible to the closely related virus Tomato yellow leaf curl Sardinia virus (TYLCSV).	

(ii) White fly inoculation method

1.	Pathogen	Tomato yellow leaf curl virus (TYLCV) IL strain
2.	Quarantine status	yes (see 13.)
3.	Host species	<i>Solanum lycopersicum</i>
4.	Source of inoculum	Spain <sup>18</sup>
5.	Isolate	TYLCV-IL La Mayora
8.	Multiplication inoculum	White flies
8.6	Harvest of inoculums	
9.	Format of the test	
9.1	Number of plants per genotype	20
9.2	Number of replicates	Two replicates
9.3	Control varieties	
	Resistant	TY 20, Anastasia, Mohawk
	Susceptible	Big Power, ( <i>Solanum lycopersicum</i> ) Moneymaker, Marmande
	Resistant	( <i>Solanum lycopersicum</i> ) Delyca, Montenegro, Anastasia, TY20, Mohawk
9.5	Test facility	greenhouse/plastic tunnel
9.9	Special measures	prevent spread of white-flies
10.	Inoculation	
10.3	Plant stage at inoculation	2-4 weeks
10.4	Inoculation method	vector (Bemisia white-flies carrying TYLCV-IL)
10.7	Final observations	1-2 months after inoculation
11.	Observations	
11.1	Method	visual
11.2	Observation scale	Symptoms: leaf yellowing and curling
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	
	absent.....[1]	severe symptoms
	present..... [9]	no or mild symptoms
13.	Critical control points: TYLCV is endemic in many tropical and subtropical areas and has a quarantine status in many countries with a temperate climate. TYLCV-IL is the strain most widely spread worldwide. With this strain, symptoms do not appear in varieties with Ty-1 and Ty-2. Some TYLCV resistant varieties may be susceptible to the closely related virus Tomato yellow leaf curl Sardinia virus (TYLCSV).	

<sup>18</sup> IHSM-UMA-CSIC, guillamon@eelm.csic.es or INIA, resistencias@inia.es

Ad. 31: Resistance to Tomato spotted wilt virus (TSWV)

Resistance to be tested in a bio-assay (method i) or in a DNA marker test (method ii), if appropriate.

(i) Bio-assay

1.	Pathogen	Tomato spotted wilt virus (see note below)
2.	Quarantine status	yes (see note below)
3.	Host species	<i>Solanum lycopersicum</i>
4.	Source of inoculum	Naktuinbouw <sup>19</sup> (NL), GEVES <sup>20</sup> (FR)
5.	Isolate	race 0, preferably a thrips-transmission deficient variant
7.	Establishment pathogenicity	biotest
8.	Multiplication inoculum	
8.6	Harvest of inoculum	symptomatic leaves may be stored at -70°C
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	Big Power and ( <i>Solanum lycopersicum</i> ) Monalbo, Momor, Montfavet H 63.5
	Resistant	Enpower and ( <i>Solanum lycopersicum</i> ) Tsunami, Bodar, Mospomor, Lisboa
9.5	Test facility	glasshouse or climatic chamber
9.6	Temperature	20°C
9.7	Light	12 hours or longer
9.9	Special measures	prevent or combat thrips
10.	Inoculation	
10.1	Preparation inoculum	press symptomatic leaves in ice-cold buffer 0,01 M PBS, pH 7.4, with 0,01 M sodium sulfite or similar buffer option: sieve the leaf sap through double muslin
10.3	Plant stage at inoculation	one or two expanded leaves
10.4	Inoculation method	mechanical, rubbing with carborundum on cotyledons, inoculum suspension < 10° C
10.7	Final observations	7-21 days after inoculation
11.	Observations	
11.1	Method	visual
11.2	Observation scale	symptoms: top mosaic, bronzing, various malformations, necrosis
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	TSWV has a quarantine status in some countries. TSWV is transmitted by <i>Thrips tabaci</i> and Western flower thrips ( <i>Frankliniella occidentalis</i> ). Pathotype 0 is defined by its inability to break resistance in tomato varieties carrying the resistance gene Sw-5.

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<sup>20</sup> GEVES: matref@geves.fr

(ii) DNA marker test

Dominant resistance gene Sw-5 is always associated with resistance to TSWV strain 0. The presence or absence of the allele for resistance can be detected by the co-dominant marker as described in Dianese, E.C. et al (2010). Specific aspects:

1.	Pathogen	Tomato spotted wilt virus
2.	Functional gene	Sw-5b
3.	Primers	
3.1	Allele for susceptibility	Sw5-Vat1-F: 5'-ACAACATCAAACAATGTTAGCC-3' Sw5-Vat2-F: 5'-CATCAAACAATGCAGTTAGCC-3'
3.2	Allele for resistance	Sw5-Res-F: 5'-ATCAACCAATACAGCCTAACC-3
3.3	Universal reverse	Sw5-universal-R: 5'-TTTCTCCCTGCAAGTTCACC-3'
3.4	Allele specific probes	Sw5-Sus1: 5'-VIC-TACATTATGAAGGGTTAACAAG-MGB-NFQ-3' Sw5-Sus2: 5'-6FAM-ACAACAGAGGGTTAACAAGTTTtagg-BHQ1-3' Sw5-Res: 5'-TEXAS RED-TGGGCGAAAATCCCAACAAG-BHQ2-3'
4.	Format of the test	
4.1	Number of plants per genotype	at least 20 plants
4.2	Control varieties	homozygous allele 1 for susceptibility present: Emperador homozygous allele 2 for susceptibility present: ( <i>Solanum lycopersicum</i> ) Mountain Magic homozygous allele for resistance present: Enpower
6.	PCR conditions	1. Initial denaturation step 10 min 95 °C 2. 40 cycles 15 sec 95 °C and 1 min 60°C. Every cycle ends with a plate reading.
8.	Interpretation of test results	
	absent .....[1]	allele(s) for susceptibility present and allele for resistance absent
	present .....[9]	allele for resistance present (homozygous or heterozygous) In case the DNA marker test result does not confirm the declaration in the TQ, a bio-assay should be performed to observe whether the variety is resistant due to another mechanism.

Ad. 32: Resistance to *Oidium neolycopersici* (On)

- |   |  |
|---|--|
| 1. Pathogen .....   | <i>Oidium neolycopersici</i> (Powdery mildew)  |
| 3. Host species .....   | <i>Solanum lycopersicum</i>  |
| 4. Source of inoculum .....   | -  |
| 5. Isolate  | see remark under 13  |
| 7. Establishment pathogenicity .....                                    | biotest  |
| 8. Multiplication inoculum  |  |
| 8.1 Multiplication medium .....   | plant  |
| 8.3 Plant stage at inoculation .....                                    | 3 weeks  |
| 8.4 Inoculation medium.....   | water  |
| 8.5 Inoculation method .....  | see 10.4   |
| 8.6 Harvest of inoculum .....   | by washing off   |
| 8.7 Check of harvested inoculum .....                                   | check for contaminants under microscope  |
| 8.8 Shelf-life/viability inoculum .....                                 | 1-2 hours  |
| 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: .....  | ( <i>Solanum lycopersicum</i> ) Momor, Montfavet H 63.5  |
| Resistant: .....  | Multifort and ( <i>Solanum lycopersicum</i> ) Atlanta, Romiro, PI-247087   |
| 9.5 Test facility .....   | glasshouse   |
| 9.6 Temperature .....   | 20°C or 18/24°C  |
| 9.7 Light .....   | 12 hours   |
| 10. Inoculation   |  |
| 10.1 Preparation inoculum.....  | collect spores in water  |
| 10.2 Quantification inoculum .....                                      | 10 <sup>4</sup> conidia/ml   |
| 10.3 Plant stage at inoculation .....                                   | 3 weeks  |
| 10.4 Inoculation method .....   | by spraying on leaves or dredging of leaves  |
| 10.7 Final observations.....  | 7-18 days after inoculation  |
| 11. Observations  |  |
| 11.1 Method.....  | visual   |
| 11.2 Observation scale .....  | 0. no sporulation<br>1. necrotic points and sometimes locally restricted sporulation<br>2. moderate sporulation<br>3. abundant sporulation |
| 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] moderate or abundant sporulation   |
| present .....   | [9] no or restricted sporulation   |
| 13. Critical control points:  |  |
- Resistance-breaking isolates should be avoided. Resistance to *O. neolycopersici* is usually race-specific. However, as long as a differential series of tomato genotypes with well defined resistances is lacking, it will remain hard to conclude that different races of *O. neolycopersici* exist.



9. Literature

Arens P., Mansilla C., Deinum D., Cavellini L., Moretti A., Rolland S., van der Schoot H., Calvache D., Ponz F., Collonnier C., Mathis R., Smilde D., Caranta C., Vosman B., 2010. Development and evaluation of robust molecular markers linked to disease resistance in tomato for distinctness, uniformity and stability testing. *Theoretical and applied genetics*. 120(3): 655-64

Dianese, E.C. et al, 2010: Development of a locus-specific, co-dominant SCAR marker for assisted-selection of the Sw-5 (Topovirus resistance) gene cluster in a wide range of tomato accessions. *Molecular Breeding*, 25(1), pp. 133-142.

Kjellberg, L., 1973: *Sortundersökningar av tomat enligt UPOV*, Swedish University of Agricultural Sciences, Research Information Centre, Alnarp Trädgaard 162, SE.

Laterrot, H., 1990: Situation de la lutte génétique contre les parasites de la Tomate dans les pays méditerranéens, P.H.M. *Revue Horticole*, No. 303, January 1990.

International Seed Federation (ISF): Plant Diseases and Resistance  
([http://www.worldseed.org/isf/diseases\\_resistance.html](http://www.worldseed.org/isf/diseases_resistance.html))

10. Technical Questionnaire

TECHNICAL QUESTIONNAIRE	Page {x} of {y}	Reference Number:																				
		Application date: (not to be filled in by the applicant)																				
<b>TECHNICAL QUESTIONNAIRE</b> to be completed in connection with an application for plant breeders' rights																						
<p>1. Subject of the Technical Questionnaire</p> <p>Tomato Rootstocks belonging to:</p> <table style="width: 100%; border-collapse: collapse;"> <tr> <td style="width: 10%; padding: 5px;">1.1</td> <td style="width: 25%; padding: 5px;">Botanical name</td> <td style="width: 60%; padding: 5px;"><input style="width: 95%;" type="text" value="Solanum habrochaites S. Knapp &amp; D.M. Spooner"/></td> <td style="width: 5%; padding: 5px;">[...]</td> </tr> <tr> <td style="padding: 5px;">1.2</td> <td style="padding: 5px;">Botanical name</td> <td style="padding: 5px;"><input style="width: 95%;" type="text" value="Solanum lycopersicum L. x Solanum habrochaites S. Knapp &amp; D.M. Spooner"/></td> <td style="padding: 5px;">[...]</td> </tr> <tr> <td style="padding: 5px;">1.3</td> <td style="padding: 5px;">Botanical name</td> <td style="padding: 5px;"><input style="width: 95%;" type="text" value="Solanum lycopersicum L. x Solanum peruvianum (L.) Mill."/></td> <td style="padding: 5px;">[...]</td> </tr> <tr> <td style="padding: 5px;">1.4</td> <td style="padding: 5px;">Botanical name</td> <td style="padding: 5px;"><input style="width: 95%;" type="text" value="Solanum lycopersicum L. x Solanum cheesmaniae (L. Ridley) Fosberg"/></td> <td style="padding: 5px;">[...]</td> </tr> <tr> <td style="padding: 5px;">1.5</td> <td style="padding: 5px;">Botanical name</td> <td style="padding: 5px;"><input style="width: 95%;" type="text" value="Solanum pimpinellifolium L. x Solanum habrochaites S. Knapp &amp; D.M. Spooner"/></td> <td style="padding: 5px;">[...]</td> </tr> </table>			1.1	Botanical name	<input style="width: 95%;" type="text" value="Solanum habrochaites S. Knapp &amp; D.M. Spooner"/>	[...]	1.2	Botanical name	<input style="width: 95%;" type="text" value="Solanum lycopersicum L. x Solanum habrochaites S. Knapp &amp; D.M. Spooner"/>	[...]	1.3	Botanical name	<input style="width: 95%;" type="text" value="Solanum lycopersicum L. x Solanum peruvianum (L.) Mill."/>	[...]	1.4	Botanical name	<input style="width: 95%;" type="text" value="Solanum lycopersicum L. x Solanum cheesmaniae (L. Ridley) Fosberg"/>	[...]	1.5	Botanical name	<input style="width: 95%;" type="text" value="Solanum pimpinellifolium L. x Solanum habrochaites S. Knapp &amp; D.M. Spooner"/>	[...]
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<p>2. Applicant</p> <table style="width: 100%; border-collapse: collapse;"> <tr> <td style="width: 30%; padding: 5px;">Name</td> <td style="padding: 5px;"><input style="width: 95%;" type="text"/></td> </tr> <tr> <td style="padding: 5px;">Address</td> <td style="padding: 5px;"><input style="width: 95%; height: 80px;" type="text"/></td> </tr> <tr> <td style="padding: 5px;">Telephone No.</td> <td style="padding: 5px;"><input style="width: 95%;" type="text"/></td> </tr> <tr> <td style="padding: 5px;">Fax No.</td> <td style="padding: 5px;"><input style="width: 95%;" type="text"/></td> </tr> <tr> <td style="padding: 5px;">E-mail address</td> <td style="padding: 5px;"><input style="width: 95%;" type="text"/></td> </tr> <tr> <td style="padding: 5px;">Breeder (if different from applicant)</td> <td style="padding: 5px;"><input style="width: 95%;" type="text"/></td> </tr> </table>			Name	<input style="width: 95%;" type="text"/>	Address	<input style="width: 95%; height: 80px;" type="text"/>	Telephone No.	<input style="width: 95%;" type="text"/>	Fax No.	<input style="width: 95%;" type="text"/>	E-mail address	<input style="width: 95%;" type="text"/>	Breeder (if different from applicant)	<input style="width: 95%;" type="text"/>								
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E-mail address	<input style="width: 95%;" type="text"/>																					
Breeder (if different from applicant)	<input style="width: 95%;" type="text"/>																					
<p>3. Proposed denomination and breeder's reference</p> <table style="width: 100%; border-collapse: collapse;"> <tr> <td style="width: 30%; padding: 5px;">Proposed denomination (if available)</td> <td style="padding: 5px;"><input style="width: 95%;" type="text"/></td> </tr> <tr> <td style="padding: 5px;">Breeder's reference</td> <td style="padding: 5px;"><input style="width: 95%;" type="text"/></td> </tr> </table>			Proposed denomination (if available)	<input style="width: 95%;" type="text"/>	Breeder's reference	<input style="width: 95%;" type="text"/>																
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#4. Information on the breeding scheme and propagation of the variety

4.1 Breeding scheme

- |       |                                |     |
|-------|--------------------------------|-----|
| (i)   | Inbred line                    | [ ] |
| (ii)  | Hybrid                         | [ ] |
| (iii) | Open-pollinated variety        | [ ] |
| (iv)  | Other (please provide details) | [ ] |

Variety resulting from:

4.1.1 Crossing

- |     |   |     |
|-----|---|-----|
| (a) | controlled cross<br>(please state parent varieties) | [ ] |
|-----|---|-----|

(.....)	x	(.....)
female parent		male parent

- |     |   |     |
|-----|---|-----|
| (b) | partially known cross<br>(please state known parent variety(ies)) | [ ] |
|-----|---|-----|

(.....)	x	(.....)
female parent		male parent

- |     |               |     |
|-----|---------------|-----|
| (c) | unknown cross | [ ] |
|-----|---------------|-----|

- |       |   |     |
|-------|---|-----|
| 4.1.2 | Mutation<br>(please state parent variety) | [ ] |
|-------|---|-----|

- |       |   |     |
|-------|---|-----|
| 4.1.3 | Discovery and development<br>(please state where and when discovered and how developed) | [ ] |
|-------|---|-----|

- |       |                                   |     |
|-------|-----------------------------------|-----|
| 4.1.4 | Other<br>(please provide details) | [ ] |
|-------|-----------------------------------|-----|

# Authorities may allow certain of this information to be provided in a confidential section of the Technical Questionnaire.

TECHNICAL QUESTIONNAIRE	Page {x} of {y}	Reference Number:
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4.2 Method of propagating the variety

4.2.1 Seed-propagated varieties

- (a) Self-pollination [ ]
- (b) Cross-pollination
  - (i) population [ ]
  - (ii) synthetic variety [ ]
- (c) Hybrid [ ]
- (d) Other [ ]  
(please provide details)

[ ]

4.2.2 Vegetatively propagated varieties

- (a) cuttings [ ]
- (b) *in vitro* propagation [ ]
- (c) other (state method) [ ]

[ ]

4.2.3 Other [ ]  
(please provide details)

[ ]

TECHNICAL QUESTIONNAIRE	Page {x} of {y}	Reference Number:
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5. Characteristics of the variety to be indicated (the number in brackets refers to the corresponding characteristic in Test Guidelines; please mark the note which best corresponds).

	Characteristics	Example Varieties	Note
<b>5.1</b>	<b>Fruit: green shoulder</b>		
<b>(11)</b>			
	absent		1[ ]
	present	Big Force, Maxifort	9[ ]
<b>5.2</b>	<b>Fruit: shape in longitudinal section</b>		
<b>(17)</b>			
	broad oblate	He-Wolf	1[ ]
	narrow oblate	Gladiator	2[ ]
	circular	Maxifort	3[ ]
	obovate		4[ ]
<b>5.3</b>	<b>Fruit: number of locules</b>		
<b>(18)</b>			
	only two	Maxifort	1[ ]
	two and three		2[ ]
<b>5.4</b>	<b>Fruit: color at maturity</b>		
<b>(19)</b>			
	green	Big Force	1[ ]
	yellowish	Vigomax	2[ ]
	orangish	Titron	3[ ]
	reddish	Brigeor	4[ ]
<b>5.5</b>	<b>Resistance to <i>Meloidogyne incognita</i> (Mi)</b>		
<b>(22)</b>			
	susceptible	Bruce	1[ ]
	moderately resistant		2[ ]
	highly resistant	Emperador	3[ ]
<b>5.6</b>	<b>Resistance to <i>Verticillium</i> sp. (Va and Vd) - Race 0</b>		
<b>(23)</b>			
	absent		1[ ]
	present	Big Power	9[ ]

TECHNICAL QUESTIONNAIRE	Page {x} of {y}	Reference Number:
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	Characteristics	Example Varieties	Note
<b>5.7</b> <b>(24)</b>	<b>Resistance to <i>Fusarium oxysporum</i> f. sp. <i>lycopersici</i> (Fol)</b>		
<b>5.8</b> <b>(24.1)</b>	<b>Race 0EU/1US</b>		
	absent		1[ ]
	present	Emperador	9[ ]
<b>5.9</b> <b>(24.2)</b>	<b>Race 1EU/2US</b>		
	absent		1[ ]
	present	Emperador	9[ ]
<b>5.10</b> <b>(24.3)</b>	<b>Race 2EU/3US</b>		
	absent	Emperador	1[ ]
	present	Colosus	9[ ]
<b>5.11</b> <b>(25)</b>	<b>Resistance to <i>Fusarium oxysporum</i> f. sp. <i>radicis-lycopersici</i> (Forl)</b>		
	absent	Kemerit	1[ ]
	present	Emperador	9[ ]

TECHNICAL QUESTIONNAIRE	Page {x} of {y}	Reference Number:
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6. Similar varieties and differences from these varieties

*Please use the following table and box for comments to provide information on how your candidate variety differs from the variety (or varieties) which, to the best of your knowledge, is (or are) most similar. This information may help the examination authority to conduct its examination of distinctness in a more efficient way.*

Denomination(s) of variety(ies) similar to your candidate variety	Characteristic(s) in which your candidate variety differs from the similar variety(ies)	Describe the expression of the characteristic(s) for the <b>similar</b> variety(ies)	Describe the expression of the characteristic(s) for <b>your</b> candidate variety
<i>Example</i>	<i>Fruit: green shoulder</i>	<i>present</i>	<i>absent</i>

Comments:

TECHNICAL QUESTIONNAIRE	Page {x} of {y}	Reference Number:
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#7. Additional information which may help in the examination of the variety

7.1 In addition to the information provided in sections 5 and 6, are there any additional characteristics which may help to distinguish the variety?

Yes [ ] No [ ]

(If yes, please provide details)

7.2 Are there any special conditions for growing the variety or conducting the examination?

Yes [ ] No [ ]

(If yes, please provide details)

7.3 Other information

8. Authorization for release

(a) Does the variety require prior authorization for release under legislation concerning the protection of the environment, human and animal health?

Yes [ ] No [ ]

(b) Has such authorization been obtained?

Yes [ ] No [ ]

If the answer to (b) is yes, please attach a copy of the authorization.



TECHNICAL QUESTIONNAIRE	Page {x} of {y}	Reference Number:
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9. Information on plant material to be examined or submitted for examination.

9.1 The expression of a characteristic or several characteristics of a variety may be affected by factors, such as pests and disease, chemical treatment (e.g. growth retardants or pesticides), effects of tissue culture, different rootstocks, scions taken from different growth phases of a tree, etc.

9.2 The plant material should not have undergone any treatment which would affect the expression of the characteristics of the variety, unless the competent authorities allow or request such treatment. If the plant material has undergone such treatment, full details of the treatment must be given. In this respect, please indicate below, to the best of your knowledge, if the plant material to be examined has been subjected to:

- |   |         |        |
|---|---------|--------|
| (a) Microorganisms (e.g. virus, bacteria, phytoplasma)    | Yes [ ] | No [ ] |
| (b) Chemical treatment (e.g. growth retardant, pesticide) | Yes [ ] | No [ ] |
| (c) Tissue culture  | Yes [ ] | No [ ] |
| (d) Other factors   | Yes [ ] | No [ ] |

Please provide details for where you have indicated "yes".

.....

10. I hereby declare that, to the best of my knowledge, the information provided in this form is correct:

Applicant's name

Signature

Date

[End of document]