



PROTOCOL FOR TESTS ON DISTINCTNESS, UNIFORMITY AND STABILITY

Capsicum annuum L.

SWEET PEPPER, HOT PEPPER, PAPRIKA, CHILI

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TABLE OF CONTENTS

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1.	SUBJECT OF THE PROTOCOL AND REPORTING	3
1.1	Scope of the technical protocol.....	3
1.2	Entry into Force	3
1.3	Reporting between Examination Office and CPVO and Liaison with Applicant	3
2.	MATERIAL REQUIRED	3
2.1	Plant material requirements	3
2.2	Informing the applicant of plant material requirements.....	4
2.3	Informing about problems on the submission of material	4
3.	METHOD OF EXAMINATION.....	4
3.1	Number of growing cycles.....	4
3.2	Testing Place	4
3.3	Conditions for Conducting the Examination.....	4
3.4	Test design.....	4
3.5	Special tests for additional characteristics.....	4
3.6	Constitution and maintenance of a variety collection	4
4.	ASSESSMENT OF DISTINCTNESS, UNIFORMITY AND STABILITY	5
4.1	Distinctness	6
4.2	Uniformity	7
4.3	Stability.....	7
5.	GROUPING OF VARIETIES AND ORGANISATION OF THE GROWING TRIAL.....	7
6.	INTRODUCTION TO THE TABLE OF CHARACTERISTICS	8
6.1	Characteristics to be used	8
6.2.	States of expression and corresponding notes.....	8
6.3	Example Varieties.....	8
6.4	Legend.....	8
7.	TABLE OF CHARACTERISTICS.....	9
8.	EXPLANATIONS ON THE TABLE OF CHARACTERISTICS.....	28
8.1	Explanations covering several characteristics	28
8.2	Explanations for individual characteristics	28
9.	LITERATURE	52
10.	TECHNICAL QUESTIONNAIRE	54

1. SUBJECT OF THE PROTOCOL AND REPORTING

1.1 Scope of the technical protocol

This Technical Protocol applies to all varieties of *Capsicum annuum* L.

The protocol describes the technical procedures to be followed in order to meet the requirements of Council Regulation 2100/94 on Community Plant Variety Rights. The technical procedures have been agreed by the Administrative Council and are based on documents agreed by the International Union for the Protection of New Varieties of Plants (UPOV), such as the General Introduction to DUS (UPOV Document TG/1/3 http://www.upov.int/export/sites/upov/resource/en/tg_1_3.pdf), its associated TGP documents (<http://www.upov.int/tgp/en/>) and the relevant UPOV Test Guideline TG/76/9 dated 09/08/2024 (<https://www.upov.int/edocs/tgdocs/en/tg076.pdf>) for the conduct of tests for Distinctness, Uniformity and Stability.

1.2 Entry into Force

The present protocol enters into force on **01.04.2025**. Any ongoing DUS examination of candidate varieties started before the aforesaid date will not be affected by the approval of the Technical Protocol. Technical examinations of candidate varieties are carried out according to the TP in force when the DUS test starts. The starting date of a DUS examination is considered to be the due date for submitting of plant material for the first growing cycle.

In cases where the Office requests to take-over a DUS report for which the technical examination has either been finalized or which is in the process to be carried out at the moment of this request, such report can only be accepted if the technical examination has been carried out according to the CPVO TP which was in force at the moment when the technical examination started.

1.3 Reporting between Examination Office and CPVO and Liaison with Applicant

1.3.1 Reporting between Examination Office and CPVO

The Examination Office shall deliver to the CPVO a preliminary report ("the preliminary report") no later than four weeks after the date of the request for technical examination by the CPVO and in any case preferably before the submission period of the plant material.

The Examination Office shall also deliver to the CPVO a report relating to each growing period ("the interim report") and, when the Examination Office considers the results of the technical examination to be adequate to evaluate the variety or the CPVO so requests, a report relating to the examination ("the final report").

The final report shall state the opinion of the Examination Office on the distinctness, uniformity and stability of the variety. Where it considers those criteria to be satisfied, or where the CPVO so requests, a description of the variety shall be added to the report.

If a report is negative the Examination Office shall set out the detailed reasons for its findings.

The interim and the final reports shall be delivered to the CPVO as soon as possible and no later than on the deadlines as laid down in the designation agreement.

1.3.2 Informing on problems in the DUS test

In cases where the Examination Office identifies issues during the course of the technical examination that may lead to a negative report, the Examination Office shall inform the CPVO and in urgent cases the applicant/holder as soon as such issues become obvious.

1.3.3 Sample keeping in case of problems

As far as feasible the Examination Office shall keep a representative sample of any relevant testing material of the candidate variety and reference variety(ies) if the technical examination has resulted in a negative report. As soon as possible, the CPVO shall inform the Examination Office when the material can be destroyed.

2. MATERIAL REQUIRED

2.1 Plant material requirements

Information with respect to the agreed closing dates and submission requirements of plant material for the technical examination of varieties can be found on <https://public.plantvarieties.eu/publication> in the special issue S2/S3 of the Official Gazette of the Office. General requirements on submission of samples are also to be found following the same link.

2.2 Informing the applicant of plant material requirements

The CPVO informs the applicant that:

- he/she is responsible for ensuring compliance with any customs and plant health requirements;
- the plant material supplied should be visibly healthy, not lacking in vigour, nor affected by any important pest or disease;
- 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.

2.3 Informing about problems on the submission of material

The Examination Office shall report to the CPVO immediately in cases where the test material of the candidate variety has not arrived in time or in cases where the material submitted does not fulfil the conditions laid down in the request for submission of plant material issued by the CPVO.

In cases where the examination office encounters difficulties to obtain plant material of reference varieties the CPVO should be informed in writing.

3. METHOD OF EXAMINATION

3.1 Number of growing cycles

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

3.1.2 The two independent growing cycles should be in the form of two separate plantings.

3.1.3 The testing of a variety may be concluded when the competent authority can determine with certainty the outcome of the test.

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" http://www.upov.int/edocs/tgpdocs/en/tgp_9.pdf.

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 2 replicates.

3.4.2 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 Special tests for additional characteristics

In accordance with Article 23 of Implementing Rules N° 874/2009 an applicant may claim either in the Technical Questionnaire or during the test that a candidate has a characteristic which would be helpful in establishing distinctness. If such a claim is made and is supported by reliable technical data, a special test may be undertaken providing that a technically acceptable test procedure can be devised.

Special tests will be undertaken, with the agreement of the President of CPVO, where distinctness is unlikely to be shown using the characteristics listed in the protocol.

3.6 Constitution and maintenance of a variety collection

The process for the constitution and the maintenance of a variety collection can be summarized as follows:

Step 1: Making an inventory of the varieties of common knowledge.

Step 2: Establishing a collection ("variety collection") of varieties of common knowledge which are relevant for the examination of distinctness of candidate varieties.

Step 3: Selecting the varieties from the variety collection which need to be included in the growing trial or other tests for the examination of distinctness of a particular candidate variety.

3.6.1 Forms of variety collection

(a) Seed propagated varieties

The variety collection shall comprise variety descriptions and living plant material, thus a living reference collection. The variety description shall be produced by the EO unless special cooperation exists between EOs and the CPVO. The descriptive and pictorial information produced by the EO shall be held and maintained in a form of a database.

(b) Vegetatively propagated varieties

The variety collection shall comprise variety descriptions; no living reference collection is required. The variety description shall be produced by the EO unless special cooperation exists between EOs and the CPVO. The descriptive and pictorial information produced by the EO shall be held and maintained in a form of a database.

3.6.2 Living Plant Material

(a) Seed propagated varieties

The EO shall collect and maintain living plant material of varieties of the species concerned in the variety collection.

(b) vegetatively propagated varieties

The EO may only obtain living plant material of reference varieties as and when those varieties need to be included in growing trials or other tests.

Living plant material of reference varieties identified to be included in the growing trial may be taken from the EO's collection in case there is one or shall be obtained specifically for the growing trial or other tests.

3.6.3 Range of the variety collection

The living variety collection shall cover at least those common knowledge varieties that are suitable to grow in the climatic conditions of a respective EO.

3.6.4 Making an inventory of varieties of common knowledge for inclusion in the variety collection

The inventory shall include varieties protected under National and Community PBR, varieties registered in the Common Catalogue, the OECD list, the Conservation variety list and varieties in trade or in commercial registers for those species not covered by a National or the Common Catalogue.

The inventory shall take into account the list of varieties which are the subject of an on-going application for protection or official registration (candidate varieties).

3.6.5 Maintenance and renewal/update of a living variety collection

(a) Seed propagated varieties

The EO shall maintain seeds in conditions which will ensure germination and viability, periodical checks, and renewal as required.

Living material in variety collections representing varieties for which a DUS test was carried out at that EO shall be renewed after verification in a side-by-side comparison. In case where no living material is available anymore in the collection, such verification could be done with any other test that has proven to give similar results between the material in the collection and the new material.

(b) Vegetatively propagated varieties

The EO shall maintain the variety collection under appropriate growing conditions (e.g., glasshouse, orchard, in vitro), where it shall be ensured that the plants are adequately irrigated, fertilised, pruned and protected from harmful pests and diseases.

Living material in variety collections representing varieties for which a DUS test was carried out at that EO shall be renewed after verification in a side-by-side comparison. In case where no living material is available anymore in the collection, such verification could be done with any other test that has proven to give similar results between the material in the collection and the new material.

4. **ASSESSMENT OF DISTINCTNESS, UNIFORMITY AND STABILITY**

The prescribed procedure is to assess distinctness, uniformity and stability in a growing trial.

4.1 Distinctness

4.1.1 General recommendations

It is of particular importance for users of this Technical Protocol to consult the UPOV-General Introduction to DUS (link in chapter 1 of this document) and TGP 9 'Examining Distinctness' (http://www.upov.int/edocs/tgpdocs/en/tgp_9.pdf) prior to making decisions regarding distinctness. However, the following points are provided for elaboration or emphasis in this Technical Protocol.

Further guidance is provided in documents TGP/9 "Examining Distinctness" and TGP/8 "Trial Design and Techniques Used in the Examination of Distinctness, Uniformity and Stability".

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 Technical Protocols are familiar with the recommendations contained in the UPOV-General Introduction to DUS 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.

For testing the resistance to certain pathogens, unless otherwise indicated, the test should be performed on at least 20 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 third 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., colour 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 this Technical Protocol to consult the UPOV-General Introduction to DUS (link in chapter 1 of this document) and TGP 10 'Examining Uniformity' (http://www.upov.int/edocs/tgpdocs/en/tgp_10.pdf) prior to making decisions regarding uniformity. However, the following points are provided for elaboration or emphasis in this Technical Protocol:
- 4.2.2 This Technical Protocol has been developed for the examination of seed-propagated varieties and vegetatively propagated varieties. For varieties with other types of propagation the recommendations in the UPOV-General Introduction to DUS and document TGP/13 "Guidance for new types and species", Section 4.5 "Testing Uniformity" should be followed.
- 4.2.3 The assessment of uniformity should be according to the recommendations for cross-pollinated varieties in the UPOV-General Introduction to DUS.
- 4.2.4 For the assessment of uniformity of self-pollinated varieties, hybrids and vegetatively propagated varieties, 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 It is of particular importance for users of this Technical Protocol to consult the UPOV-General Introduction to DUS (link in chapter 1 of this document) and TGP 11 'Examining Stability' (http://www.upov.int/edocs/tgpdocs/en/tgp_11.pdf)

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.

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 ORGANISATION 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 organise the growing trial so that similar varieties are grouped together.
- 5.3 The following have been agreed as useful grouping characteristics:
- a) Plant: shortened internodes (characteristic 4)
 - b) Flower: anthocyanin coloration of anther (characteristic 23)
 - c) Immature fruit: colour (characteristic 26)
 - d) Fruit: length (characteristic 30)
 - e) Fruit: diameter (characteristic 31)
 - f) Fruit: ratio length/diameter (characteristic 32)
 - g) Fruit: shape in longitudinal section (characteristic 33)
 - h) Fruit: colour (characteristic 41)
 - i) Fruit: capsaicin in placenta (characteristic 48)
 - j) Resistance to Tobamovirus - *Tobacco mosaic virus* - Group 0 (TMV: 0) (characteristic 54)
 - k) Resistance to Tobamovirus - *Pepper mild mottle virus* - Group 2 (PMMoV: 1.2) (characteristic 55)
 - l) Resistance to Tobamovirus - *Pepper mild mottle virus* - Group 3 (PMMoV: 1.2.3) (characteristic 56)
 - m) Resistance to *Potato Y virus* (PVY) - Pathotype 0 (PVY: 0) (characteristic 57)
 - n) Resistance to *Tomato spotted wilt virus* Pathotype 0 (TSWV: 0) (characteristic 62)

- 5.4** If characteristics other than those mentioned in the list of grouping characteristics and/or from the table of characteristics and/or from the Technical Questionnaire – sections 5 and 7. are used for the selection of varieties to be included into the growing trial, the EO shall inform the CPVO and seek the prior consent of the CPVO before using these characteristics.
- 5.5** Guidance for the use of grouping characteristics, in the process of examining distinctness, is provided through the UPOV-General Introduction to DUS and document TGP/9 “Examining Distinctness”.

6. INTRODUCTION TO THE TABLE OF CHARACTERISTICS

6.1 Characteristics to be used

The characteristics to be used in DUS tests and preparation of descriptions shall be those referred to in the table of characteristics. All the characteristics shall be used, providing that observation of a characteristic is not rendered impossible by the expression of any other characteristic, or the expression of a characteristic is prevented by the environmental conditions under which the test is conducted or by specific legislation on plant health. In the latter case, the CPVO should be informed.

The Administrative Council empowers the President, in accordance with Article 23 of Commission Regulation N°874/2009, to insert additional characteristics and their expressions in respect of a variety.

Asterisked characteristics

In the case of disease resistance characteristics, only those resistances marked with an asterisk (*) in the CPVO column are compulsory.

6.2. States of expression and corresponding notes

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. All relevant states of expression are presented in the characteristic.

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

6.3 Example Varieties

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

6.4 Legend

For column ‘CPVO N°’:

G	Grouping characteristic	-see Chapter 5
QL	Qualitative characteristic	
QN	Quantitative characteristic	
PQ	Pseudo-qualitative characteristic	
(+)	Explanations for individual characteristics	-see Chapter 8.2
(*)	Asterisked characteristic	-see Chapter 6.1

For column ‘UPOV N°’:

The numbering of the characteristics is provided as a reference to the UPOV guideline.

(*)	UPOV Asterisked characteristic	- Characteristics that are important for the international harmonization of variety descriptions.
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For column ‘Stage, method’:

MG, MS, VG, VS		-see Chapter 4.1.5
(a)-(d)	Explanations covering several Characteristics	-see Chapter 8.1

7. TABLE OF CHARACTERISTICS

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note	
1. QL	1.	VG	Seedling: anthocyanin coloration of hypocotyl			
			absent	Albaregia	1	
			present	Lamuyo	9	
2. (+) QN	2.	VG	Plant: habit			
			(a)	upright	De Cayenne, Doux très long des Landes, Piquant d'Algérie	1
				semi-upright	Sonar	2
			prostrate		3	
3. (+) QN	3. (*)	MG/MS /VG	Plant: height			
			(a)	very short		1
				very short to short		2
				short	Bravia	3
				short to medium		4
				medium	HRF	5
				medium to tall		6
				tall	Century	7
				tall to very tall		8
	very tall	Brutus	9			
4. (+) QL G	4. (*)	VG	Plant: shortened internodes			
			(a)	absent	California wonder, De Cayenne	1
			present	Bucano	9	

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note		
5. (+)	5.	MS	<u>Only varieties with plant:</u> <u>shortened internodes: present:</u> number of internodes between the first flower and shortened internodes				
			QN	(a)	none	1	
					one to three	2	
					more than three	3	
6. (+)	6.	MS/VG	<u>Only varieties with plant:</u> <u>shortened internodes: absent:</u> length of internodes				
			QN	(a)	very short	Albaregia	1
					very short to short		2
					short	Tenor	3
					short to medium		4
					medium	Florian	5
					medium to long		6
					long	Corno di toro rosso	7
					long to very long		8
					very long	Fenice	9
7. (+)	7.	MS/VG	Stem: length				
			QN	(a)	very short		1
					very short to short		2
					short	Bomenta, Corvinus	3
					short to medium		4
					medium	Bravia, Lamuyo, Nestoss, Remus	5
					medium to long		6
					long	Lipari, Marconi	7
					long to very long		8
very long		9					

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note
8.	8.	VG	Stem: intensity of anthocyanin coloration of nodes		
QN	(a)		absent or very weak	Bravia, Nestoss, Remus	1
			very weak to weak		2
			weak	California wonder	3
			weak to medium		4
			medium	Lamuyo, Sonar	5
			medium to strong		6
			strong	Piquant d'Algérie	7
			strong to very strong		8
			very strong	Smolder	9
9.	9.	VG	Stem: hairiness of nodes		
QN	(a)		absent or very weak	Arlequin	1
			very weak to weak		2
			weak	Bravia, Nestoss	3
			weak to medium		4
			medium	Doux très long des Landes, Farnese	5
			medium to strong		6
			strong	Fenice, Solario	7
			strong to very strong		8
			very strong	Brutus	9

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note
10. (+)	10.	MS/VG	Leaf blade: length		
QN		(a)	very short	Macska sárga	1
			very short to short		2
			short	De Cayenne	3
			short to medium		4
			medium	Marconi	5
			medium to long		6
			long	Allrounder	7
			long to very long		8
			very long	Solario	9
11. (+)	11.	MS/VG	Leaf blade: width		
QN		(a)	very narrow	Macska sárga	1
			very narrow to narrow		2
			narrow	De Cayenne	3
			narrow to medium		4
			medium	Marconi	5
			medium to broad		6
			broad	Allrounder	7
			broad to very broad		8
			very broad	Solario	9
12. (+)	12.	VG	Leaf blade: ratio length/width		
PQ		(a)	low	Solario	1
			medium	Balico, Sonar	2
			high	Brutus, De Cayenne	3

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note
13.	13.	VG	Leaf blade: intensity of green colour		
QN		(a)	very light		1
			very light to light		2
			light	Blondy	3
			light to medium		4
			medium	Allrounder, Frazier	5
			medium to dark		6
			dark	Rioverde	7
			dark to very dark		8
			very dark	Japo, Morrón de conserva 3, Roial	9
14. (+)	14.	VG	Leaf blade: intensity of anthocyanin coloration of upper side		
QN		(a)	absent or very weak		1
			weak	Omiyamurasaki, Purple Rain	2
			medium	Calico	3
			strong	Black Pearl	4
			very strong	Purple Flash, Takiama Purple to Red, TF802	5
15. (+)	15.	VG	Leaf blade: distribution of anthocyanin coloration of lower side		
PQ		(a)	absent		1
			on veins throughout	Takiama Purple to Red	2
			on veins and diffuse on distal part		3
			on veins and diffuse throughout	Black Pearl, Purple Flash	4
			throughout	TF802	5
16. (+)	16.	VG	Leaf blade: variegation		
QL		(a)	absent	Omiyamurasaki	1
			present	Calico, Purple Rain	9

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note
17. QN	17.	VG (a)	Leaf blade: undulation of margin		
			absent of very weak	De Cayenne	1
			very weak to weak		2
			weak	Doux très long des Landes	3
			weak to medium		4
			medium	Tenor	5
			medium to strong		6
			strong	Tosca	7
			strong to very strong		8
		very strong		9	
18. QN	18.	VG (a)	Leaf blade: blistering		
			very weak	Brutus	1
			very weak to weak		2
			weak	Pusztagold	3
			weak to medium		4
			medium	Bravia, Nestoss	5
			medium to strong		6
			strong	Greygo	7
			strong to very strong		8
	Florian	9			

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note
19. QN	19.	VG (a)	Leaf blade: glossiness		
			very weak		1
			very weak to weak		2
			weak	Brutus, Doux très long des Landes	3
			weak to medium		4
			medium	Bravia	5
			medium to strong		6
			strong	Floridor	7
			strong to very strong		8
			very strong		9
20. (+) QN	20.	MG/VG /MS	Time of beginning of flowering		
			very early		1
			very early to early		2
			early	Brutus	3
			early to medium		4
			medium	Allrounder, Lamuyo	5
			medium to late		6
			late	Piquant d'Algérie	7
			late to very late		8
			very late		9
21. (+) PQ	21.	VG (b)	Flower: attitude of pedicel		
			erect	Floridor	1
			semi-drooping	Bravia	2
			drooping	Brutus, Lamuyo	3

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note
22. PQ	22.	VG (b)	Flower: colour		
			white	Lamuyo	1
			light purple		2
			medium purple		3
			dark purple	Black Pearl	4
23 (+) QL G	23 (*)	VG (b)	Flower: anthocyanin coloration of anther		
			absent	Bravia	1
			present	Brutus, Lamuyo	9
24 (+) QL	24	VG (b)	Flower: anthocyanin coloration of filament		
			absent	AG33	1
			present	Bao-11, Morningput	9
25 (+) QN	25	VS (b)	Male sterility		
			absent	California wonder	1
			partially present		2
			totally present	Angelito	3
26 (+) PQ G	26 (*)	VG (c)	Immature fruit: colour		
			greenish white	Bravia	1
			greenish yellow	Don, Sweet banana	2
			green	Allrounder, Black Bullet, Cornus, Hitman, Impala, Syrto	3
			purple	Cardinal, Lilo, Loco, Tequila, Tonaya	4

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note
27.	27. (*)	VG	<u>Only varieties with Immature fruit:</u> <u>colour: green or purple:</u> Immature fruit: intensity of colour		
QN		(c)	very light		1
			very light to light		2
			light	Cornus, Loco, Syrto	3
			light to medium	Tequila	4
			medium	Allrounder	5
			medium to dark	Cardinal	6
			dark	Impala, Lilo, Tonaya	7
			dark to very dark		8
			very dark	Black Bullet, Hitman	9
28.	28.	VG	<u>Excluding varieties with Immature fruit:</u> <u>colour: purple:</u> Immature fruit: anthocyanin coloration		
QN		(c)	absent or weak	Lamuyo	1
			medium		2
			strong	Sweet banana	3
29. (+)	29.	VG	Fruit: attitude		
PQ		(d)	erect	Pusztagold	1
			horizontal	PAZ szentesi	2
			drooping	De Cayenne, Lamuyo	3

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note
30. (+)	30. (*)	MS/VG	Fruit: length		
QN		(d)	very short	Cherry Bomb, PAZ szentesi	1
			very short to short		2
			short	Ophelia, Smolder	3
			short to medium		4
			medium	California wonder	5
			medium to long		6
			long	Bravia, De Cayenne	7
			long to very long		8
G			very long	Carboni, Corno di toro rosso, Doux très long des Landes	9
31. (+)	31. (*)	MS/VG	Fruit: diameter		
QN		(d)	very small	De Cayenne	1
			very small to small		2
			small	Cherry Bomb	3
			small to medium		4
			medium	Doux italien	5
			medium to large		6
			large	Lamuyo, Maduro	7
			large to very large		8
G			very large	Floridor, Ibleor	9

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note
32. (+)	32. (*)	MS/VG	Fruit: ratio length/diameter		
QN		(d)	very low	Liebesapfel, PAZ szentesi	1
			very low to low		2
			low	Bucano	3
			low to medium		4
			medium	Maduro	5
			medium to high		6
			high	Lamuyo, Vidi	7
			high to very high		8
G			very high	De Cayenne, Doux très long des Landes	9
33. (+)	33. (*)	VG	Fruit: shape in longitudinal section		
PQ		(d)	oblate	Koral	1
			circular	Capperino	2
			elliptic		3
			transverse rectangular	Liebesapfel, PAZ szentesi	4
			square	Maranello	5
			rectangular	Raggio	6
			cordiform	Morrón de conserva 3	7
			ovate	Jalapeño	8
			triangular	Bravia, Corno di toro rosso, De Cayenne	9
G			trapezoid	Altea	10
34. (+)	34.	VG	Fruit: curvature		
PQ		(d)	absent	Kappy, Lamuyo	1
			C-shaped	Sweet banana	2
			S-shaped	Doux italien	3

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note
35. (+) QN	35.	VG (d)	Fruit: twisting		
			absent or weak	California wonder	1
			medium	Bubión	2
			strong	BN8707	3
36. (+) PQ	36.	VG (d)	Fruit: shape in cross section		
			elliptic	Sweet banana	1
			angular	Solario	2
			circular	Doux très long des Landes	3
37. (+) QN	37. (*)	VG (d)	Fruit: situation of pericarp at basal part		
			absent or very weak	Smolder	1
			very weak to weak		2
			weak	Donat, Kappy	3
			weak to medium		4
			medium	Banán	5
			medium to strong		6
			strong	Hawker	7
			strong to very strong		8
			very strong	Doux italien, Gelber Spiral	9
38. (+) QN	38. (*)	VG (d)	Fruit: situation of pericarp excluding basal part		
			absent or weak	Sonar, Yolo Wonder	1
			medium	Rodri	2
			strong	De Cayenne, Doux italien	3

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note
39.	39.	VG	Fruit: shape of apex		
	(*)				
PQ		(d)	strongly acute	De Cayenne	1
			moderately acute	Kappone	2
			rounded	Red Tinkerbell	3
			moderately depressed	Maduro	4
			strongly depressed	Monte	5
40.	40.	VG	Fruit: texture of surface		
(+)					
QN		(d)	smooth or weakly wrinkled	Smolder	1
			moderately wrinkled		2
			strongly wrinkled		3
41.	41.	VG	Fruit: colour		
(+)	(*)				
PQ		(d)	yellow	Allrounder	1
			orange	Arancia	2
			red	Lamuyo	3
			brown	Bastan, Chocology	4
G			green	Raymond	5
42.	42.	VG	Fruit: intensity of colour		
(+)	(*)				
QN		(d)	very light		1
			very light to light		2
			light		3
			light to medium		4
			medium		5
			medium to dark		6
			dark		7
			dark to very dark		8
			very dark		9

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note
43. QN	43.	VG (d)	Fruit: glossiness		
			very weak		1
			very weak to weak		2
			weak	Macska sárga	3
			weak to medium		4
			medium	Sonar	5
			medium to strong		6
			strong	Doux italien	7
			strong to very strong		8
	very strong	Ocelot	9		
44. QN	44. (*)	VG (d)	Fruit: depth of stalk cavity		
			absent or very shallow	Sweet banana	1
			very shallow to shallow		2
			shallow	Doux italien	3
			shallow to medium		4
			medium	Lamuyo, Maduro	5
			medium to deep		6
			deep	Baquero	7
			deep to very deep		8
	very deep	Dumbo34	9		

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note
45. (+) QN	45.	VG (d)	Fruit: depth of interloculary grooves		
			absent or very shallow	De Cayenne	1
			very shallow to shallow		2
			shallow	Kappone	3
			shallow to medium		4
			medium	Lamuyo, Marconi	5
			medium to deep		6
			deep	Round of Hungary	7
			deep to very deep		8
			very deep		9
46. QN	46. (*)	MG/VG (d)	Fruit: number of locules		
			predominantly two	De Cayenne	1
			equally two and three	Banán	2
			predominantly three	Century	3
			equally three and four	Lamuyo, Sonar	4
			predominantly four	PAZ szentesi	5
47. QN	47. (*)	VG (d)	Fruit: thickness of flesh		
			very thin	De Cayenne, Macska sárga	1
			very thin to thin		2
			thin	Banán, Doux très long des Landes	3
			thin to medium		4
			medium	Lamuyo	5
			medium to thick		6
			thick	Deimos	7
thick to very thick		8			
			very thick	Solario	9

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note		
48. (+)	48. (*)	VG/MS	Fruit: capsaicin in placenta				
			QL	(d) absent	Sonar, Sweet banana	1	
			G	present	De Cayenne	9	
49. (+)	49.	VG	Fruit: seeds				
			QL	(d) absent	Angelito	1	
				present	Lamuyo	9	
50. (+)	50.	MS/VG	Stalk: length				
			QN	(d)	very short	Jablina	1
					very short to short		2
					short	Corvinus, Yolo Wonder	3
					short to medium		4
					medium	Sonar	5
					medium to long		6
					long	De Cayenne	7
					long to very long		8
					very long	Farnese, Lipari	9
51. (+)	51.	MS/VG	Stalk: thickness				
			QN	(d)	very thin	De Cayenne, Doux très long des Landes, Macska sárga	1
					very thin to thin		2
					thin	Sweet banana	3
					thin to medium		4
					medium	Doux italien	5
					medium to thick		6
					thick	Lamuyo	7
					thick to very thick		8
					very thick		9

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note	
52. (+) QN	52.	VG	Calyx: aspect			
			(d)	non enveloping	Lamuyo, Sonar	1
				semi enveloping		2
				enveloping	De Cayenne, Sweet banana	3
53. (+) QN	53. (*)	MG/VG	Time of maturity			
				very early	Macska sárga, Madison	1
				very early to early		2
				early	Kosmik	3
				early to medium		4
				medium	Lamuyo, Sonar	5
				medium to late		6
				late	Doux d'Espagne	7
				late to very late		8
	very late	Teseo	9			
54. (+) (*) QL G	54.	VG	Resistance to Tobamovirus - <i>Tobacco mosaic virus</i> - Group 0 (TMV: 0)			
				absent	Lamu, Pepita, Piquillo	1
				present	Fehérozön, Ultron, Yolo Wonder	9
55. (+) (*) QL G	55.	VG	Resistance to Tobamovirus - <i>Pepper mild mottle virus</i> - Group 2 (PMMoV: 1.2)			
				absent	Fehérozön, Lamu, Yolo Wonder	1
				present	Achille, Candela, Ferrari, Fudji, Novi 3	9
56. (+) (*) QL G	56.	VG	Resistance to Tobamovirus - <i>Pepper mild mottle virus</i> - Group 3 (PMMoV: 1.2.3)			
				absent	Candela, Ferrari, Oida, Yolo Wonder	1
				present	Ettore, Friendly, Tom4	9

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note	
57. (+) (*) QL	57.	VG	Resistance to <i>Potato Y virus</i> (PVY) - Pathotype 0 (PVY: 0)	absent	Ferrari, Murillo, Piquillo, Yolo Wonder	1
				present	Andalus, Goleador, Vidi, Yolo Y	9
58. (+) QL	58.	VG	Resistance to <i>Potato Y virus</i> (PVY) - Pathotype 1 (PVY: 1)	absent	Yolo Wonder, Yolo Y	1
				present	Florida VR2, Ribatejo	9
59. (+) QL	59.	VG	Resistance to <i>Potato Y virus</i> (PVY) - Pathotype 1.2 (PVY: 1.2)	absent	Florida VR2, Yolo Wonder, Yolo Y	1
				present	Chouca, Serrano Criollo de Morelos 334	9
60. (+) QL	60.	VG	Resistance to <i>Phytophthora capsici</i> (Pc)	absent	Yolo Wonder	1
				present	Chistera, Favolor, Phyto 636, Solario	9
61. (+) QL	61.	VG	Resistance to <i>Cucumber mosaic virus</i> (CMV)	absent	Yolo Wonder	1
				present	Alby, Ducato, Favolor	9
62. (+) (*) QL	62.	VG	Resistance to <i>Tomato spotted wilt virus</i> Pathotype 0 (TSWV: 0)	absent	Yolo Wonder	1
				present	Galileo, Jackal, Jackpot	9
63. (+) QL	63.	VG	Resistance to <i>Xanthomonas</i> spp (ex <i>Xanthomonas campestris</i> pv. <i>vesicatoria</i>) (X spp (ex Xcv)) - Pathotype 1	absent	Yolo Wonder	1
				present	Filidor, San Marco	9

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note	
64. (+)	64.	VG	Resistance to <i>Xanthomonas</i> spp (ex <i>Xanthomonas campestris</i> pv. <i>vesicatoria</i>) (X spp (ex Xcv)) - Pathotype 2			
				QL	absent	Yolo Wonder
				present	Filidor, San Marco	9
65. (+)	65.	VG	Resistance to <i>Xanthomonas</i> spp (ex <i>Xanthomonas campestris</i> pv. <i>vesicatoria</i>) (X spp (ex Xcv)) - Pathotype 3			
				QL	absent	Yolo Wonder
				present	Filidor, San Marco	9
66. (+)	66.	MS/VG	Resistance to <i>Meloidogyne</i> <i>incognita</i> (Mi)			
				QL	absent	Tom4, Yolo Wonder
				present	Bastion, Capital, Kation, W4	9

8. EXPLANATIONS ON THE TABLE OF CHARACTERISTICS

8.1 Explanations covering several characteristics

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

- a) Observations on plant, stem, internodes and leaves should be made at the time of the first colour change of the fruit. Furthermore, observations on stem and leaves should be made at the middle third of the plant and observations on leaves should be made on fully developed leaves.
- b) Observations should be made at the middle third of the plant on fresh fully open flowers.
- c) Observations should be made before the first colour change of the fruit.
- d) Observations should be made at maturity, after the time of the colour change.

8.2 Explanations for individual characteristics

Ad. 2: Plant: habit

Observations should be made on plants without pruning or training.

Ad. 3: Plant: height

Observations should be made after a fruit set on several nodes. A poor fruit set may influence the Vigor and thus the height of the plant.

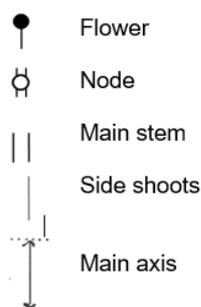
Ad. 4: Plant: shortened internodes

Observations should be made in the upper part of plants, which have not been pruned. The shoot system of pepper consists of main stems developing from the main axis, and side shoots which develop from the nodes on the main axis and on the main stems.

Absent: The main stems grow indeterminately; one or two flowers develop per node and shortened internodes never develop.

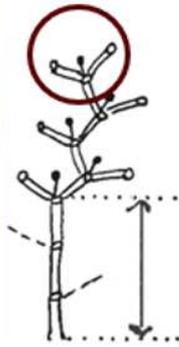
Present: After the first branching of the main axis, shorter internodes appear, and the growth of the main stem ends in a bunch of flowers.

Explanation of plant parts





1
absent

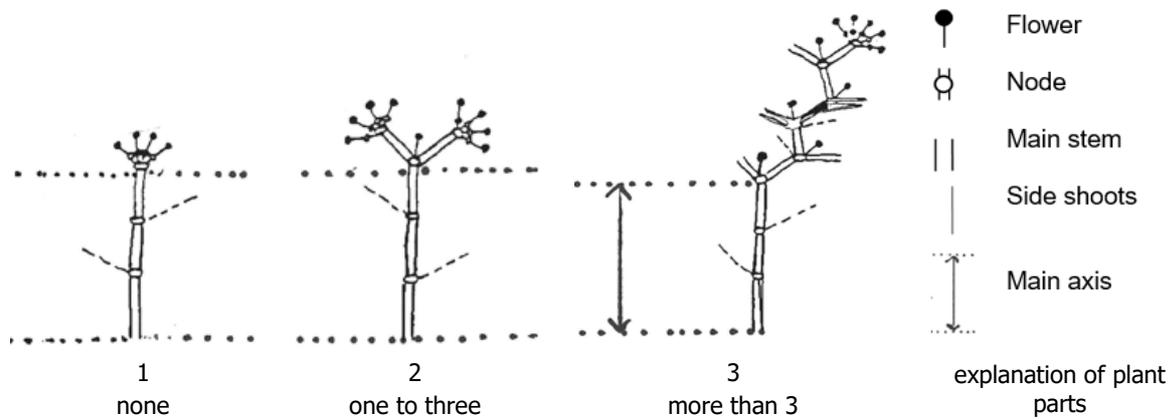


9
present



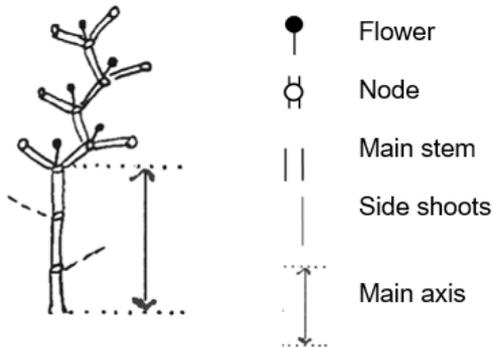
Ad. 5: Only varieties with plant: shortened internodes: present: number of internodes between the first flower and shortened internodes

Observations should be made on plants which have not been pruned, in the upper part, after the first branching of the main axis, to where the shorter internodes appear, and the main stem ends in a bunch of flowers.



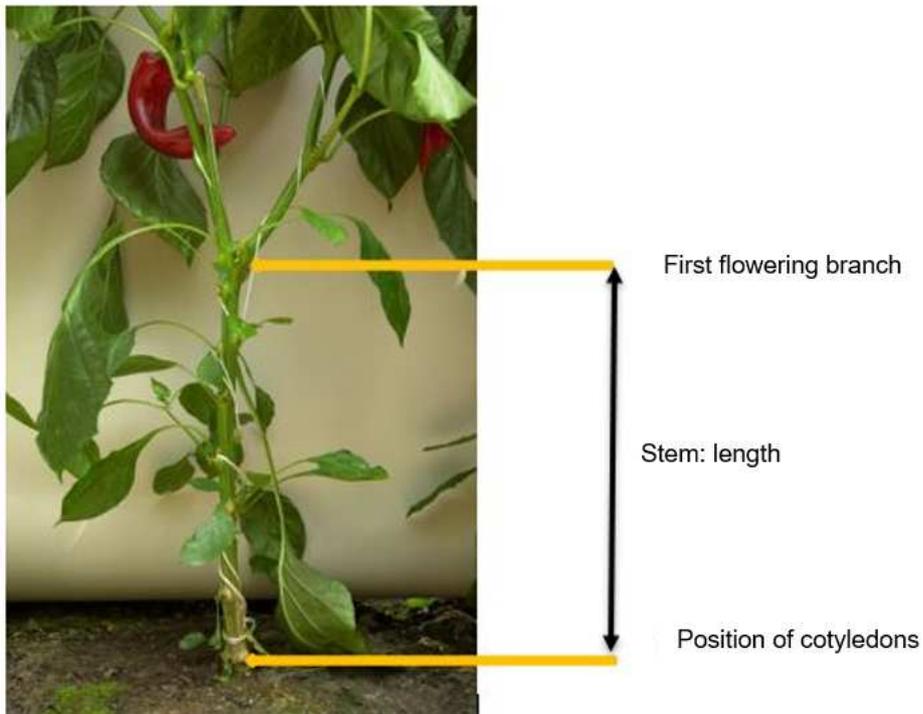
Ad. 6: Only varieties with plant: shortened internodes: absent: length of internodes

Observations should be made on plants which have not been pruned, in the upper part after the first branching of the main axis, on primary side shoots.

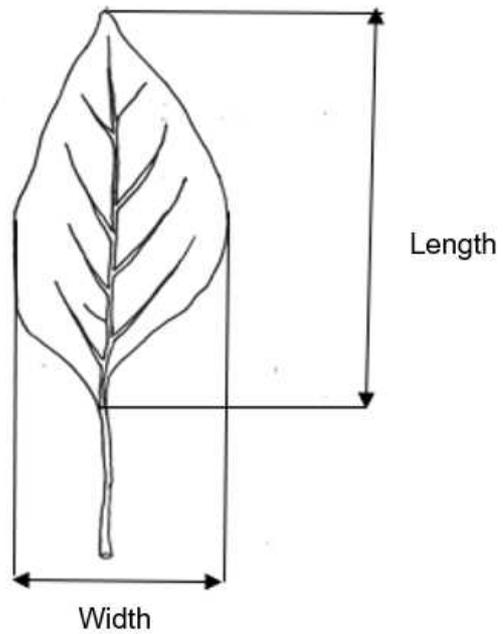


Ad. 7: Stem: length

Observations should be made from the cotyledons to the node of the first flowering branch.



Ad. 10: Leaf blade: length



Ad. 11: Leaf blade: width

See Ad. 10

Ad. 12: Leaf blade: ratio length/width

See Ad. 10

Ad. 14: Leaf blade: intensity of anthocyanin coloration of upper side

Observations should be made on recently fully developed leaves.

Ad. 15: Leaf blade: distribution of anthocyanin coloration of lower side

See Ad. 14 for time of observation.



2

on veins throughout



3

on veins and diffuse on distal part



4

on veins and diffuse throughout



5

throughout

Ad. 16: Leaf blade: variegation



9

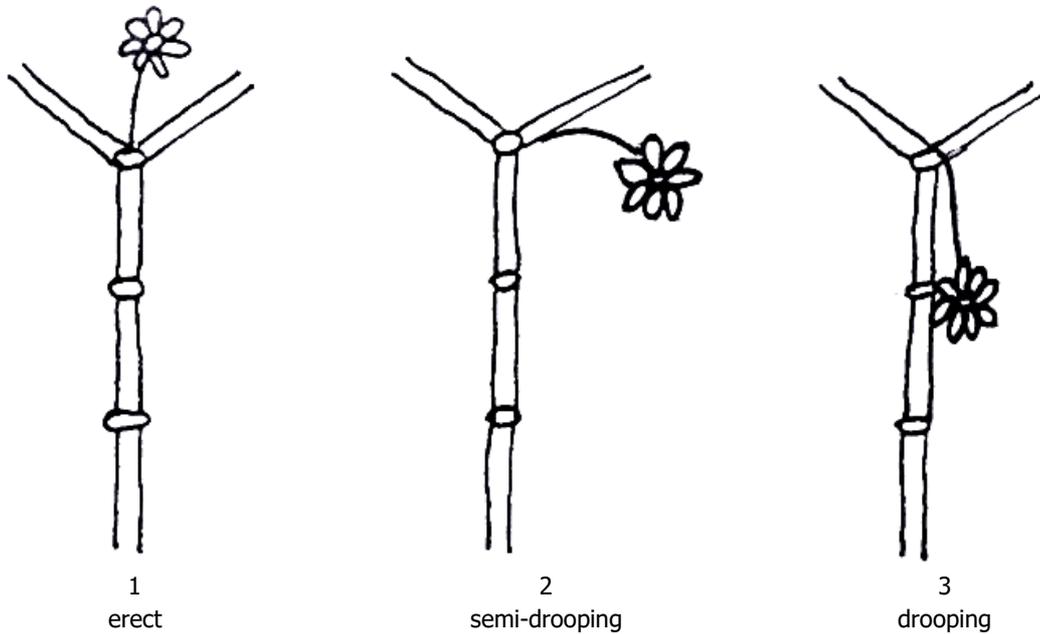
present

Ad. 20: Time of beginning of flowering

Time of beginning of flowering is reached when 50% of the plants have the first open flower of the second flowering node.

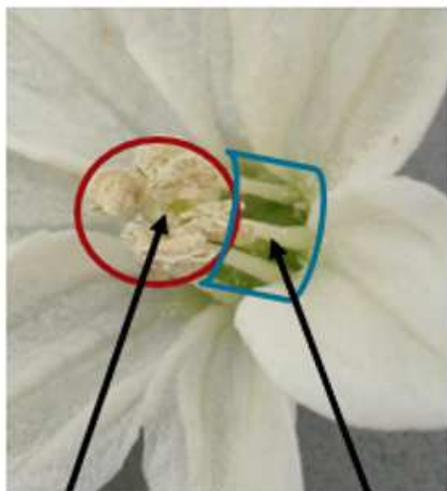
Ad. 21: Flower: attitude of pedicel

The predominant state of expression should be observed.



Ad. 23: Flower: anthocyanin coloration of anther

Observations should be made on the part of the stamen that normally produces pollen, i.e. the anther.



Anthers

Filaments

Ad. 24: Flower: anthocyanin coloration of filament

See Ad. 23

Observations should be made on the stalk of the stamen, i.e. the filament.

Ad. 25: Male sterility

Observations should be made on anthers of fresh, fully open flowers. Male sterile flowers do not have pollen.

In heterozygous genotypes, male sterility may segregate. If the segregation occurs in the predicted manner, the variety should be classified as partly present.

In hybrid production, this population is used as a mother line.



fertile



sterile

Ad. 26: Immature fruit: colour

For varieties with greenish white and greenish yellow immature fruits, particular attention is needed to make observations before the start of the colour change.

Ad. 29: Fruit: attitude

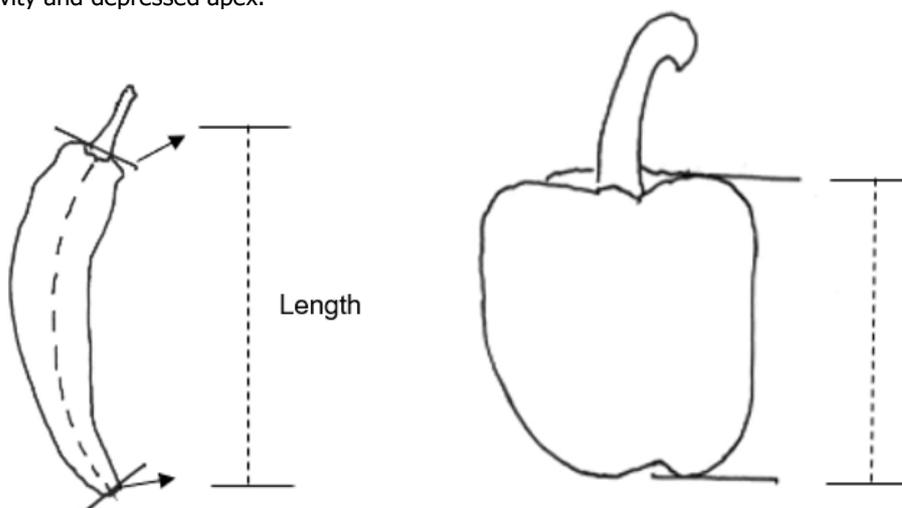
The predominant state of expression should be scored.

Ad. 30: Fruit: length

Observations of fruit length should not include the stalk.

The length of the fruit for curved or s-shaped fruits should be observed following the C- shape or S shape.

The length of the fruit with stalk cavity or/and depressed apex should be observed without taking into account the cavity and depressed apex.



Ad. 31: Fruit: diameter

Observations should be made at the broadest part of the fruit.

Ad. 32: Fruit: ratio length/diameter

ratio length/diameter	1					
	2					
	3					
	4					
	5					
	6					
	7					
	8					
	9					

Ad. 33: Fruit: shape in longitudinal section

	broadest part				
	at middle		above middle		
elongated	 3 elliptic	 6 rectangular	 8 ovate	 9 triangular	 10 trapezoid
medium ratio	 2 circular	 5 square	 7 cordiform		
compressed	 1 oblate	 4 transverse rectangular			

Ad. 34: Fruit: curvature

Observations should be made excluding the extreme point of the tip. The predominant expression should be scored.



1
absent



2
C-shaped



3
S-shaped

Ad. 35: Fruit: twisting



1
absent or weak



2
medium



3
strong

Ad. 36: Fruit: shape in cross section

Observations should be made at level of the placenta.

Ad. 37: Fruit: sinuation of pericarp at basal part



1
absent or very weak

3
weak

5
medium

7
strong

9
very strong

Ad. 38: Fruit: situation of pericarp excluding basal part



1
absent or weak



2
medium



3
strong

Ad. 40: Fruit: texture of surface



1
smooth or weakly wrinkled



2
moderately wrinkled



3
strongly wrinkled

Ad. 41: Fruit: colour

Fruit: intensity of colour (Char. 42)	Fruit: colour (Char. 41)				
	1 yellow	2 orange	3 red	4 brown	5 green
1 very light					
3 light	Deseo, Lumos, Gialte		Doyum, Healey, Teseo		
5 medium	Allrounder, Rialto, Valdor	Arancia, DSP 7054, Jack Miller	Baquero, California Wonder, Greygo, Lamuyo	Chocolony	Raymond
7 dark	Lalin, Tenor, Verdial	Delirio, Zajda	Angelito, Doux italien, Ettore		
9 very dark			Szegedi 20	Bastan	

Ad. 42: Fruit: intensity of colour

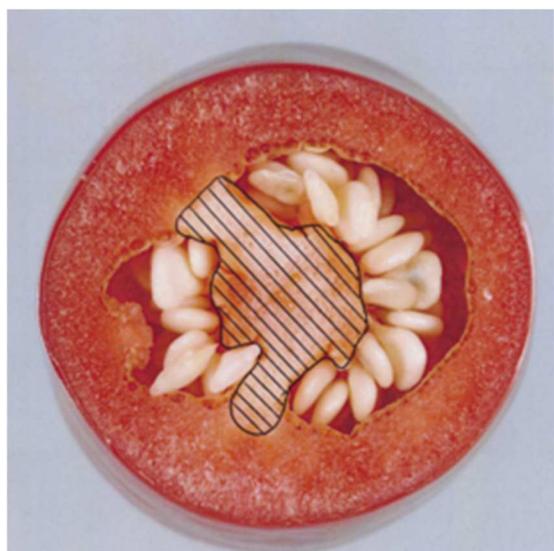
See Ad. 41 for example varieties.

Ad. 45: Fruit: depth of interlocular grooves

Observations should be made on the middle third of the fruit.

Ad. 48: Fruit: capsaicin in placenta

Observations should be made by tasting the placenta. The placenta is the tissue to which the seeds are attached to.



Ad. 49: Fruit: seeds



1
absent



9
present

Ad. 51: Stalk: thickness

Observations should be made at the middle of the stalk.

Ad. 52: Calyx: aspect

Observations should be made on whether the calyx is not enveloping (1) the fruit or enveloping the fruit including its shoulder (3), or partly enveloping the fruit, except the shoulder (2).



1
non enveloping



2
semi enveloping



3
enveloping

Ad. 53: Time of maturity

Observations should be made when at least 50% of the plants show the colour change of the fruit.

Ad. 54: Resistance to Tobamovirus - Tobacco mosaic virus- Group 0 (TMV: 0)

1.	Pathogen	Tobamovirus (the genus containing <i>Tobacco mosaic virus</i> (TMV), and <i>Pepper mild mottle virus</i> (PMMoV))
2.	Quarantine status	No
3.	Host species	Sweet pepper, hot pepper, paprika and chili – <i>Capsicum annuum</i> L.
4.	Source of inoculum	GEVES ¹ (FR), Naktuinbouw ² (NL) or INIA - CSIC ³ (SP)
5.	Isolate	- <i>Tobacco mosaic virus</i> group 0 (TMV: 0) strain Vi-6 - <i>Pepper mild mottle virus</i> group 2 (PMMoV: 1.2) strain nt203 - <i>Pepper mild mottle virus</i> group 3 (PMMoV: 1.2.3) strain Eve The test protocols have been validated in a CPVO co-funded project ⁴ with these 3 isolates/races
6.	Establishment isolate identity	genetically defined pepper differentials (ref. ISF site Feb. 2020: http://www.worldseed.org/isf/differential_hosts.html)

	Pepper Tobamovirus Group	0	1	2	3
	ISF Code →	TMV: 0,1,2 ToMV: 0,1,2 BPMoV	TMGMV PaMMV	PMMoV: 1.2	PMMoV: 1.2.3
Differential hosts	Gene				
Lamu, Early Calwonder	-	S	S	S	S
Tisana, Yolo Wonder	L1	HR	S	S	S
Tabasco	L2	HR	HR	S	S
Solario F1, Novi 3, PI159236	L3	HR	HR	HR	S
Tom4, PI260429	L4	HR	HR	HR	HR

S = susceptible; HR = highly resistant;

TMV= *Tobacco mosaic virus*; ToMV= *Tomato mosaic virus*;

PMMoV= *Pepper mild mottle virus*; TMGMV= *Tobacco mild green mosaic virus*;

BPMoV= *Bell pepper mottle virus*; PaMMV= *Paprika mild mottle virus*

7.	Establishment pathogenicity	Test on susceptible plants
8.	Multiplication inoculum	
8.1	Multiplication medium	Regeneration of the virus of plant material before inoculum preparation.
8.2	Multiplication variety	On susceptible pepper variety, Tobamovirus groups may be multiplied on varieties which are selective for each particular group. For TMV, because tomato and tobacco <i>Nicotiana tabacum</i> cv. Samsun have large leaves and can produce a lot of inoculum, they are recommended for the multiplication of TMV: 0.
8.3	Plant stage at inoculation	see 10.3
8.4	Inoculation medium	see 10.1
8.5	Inoculation method	see 10.4
8.6	Harvest of inoculum	Symptomatic fresh leaves
8.7	Check of harvested inoculum	option: on young leaves of <i>Nicotiana tabacum</i> "Xanthi", check for local lesions after 5-7 days at 20-25°C.
8.8	Shelf life/viability inoculum	fresh > 1 day in fridge, desiccated > 1 year in fridge, or juice > 1 year in freezer at - 20°C.
9.	Format of the test	
9.1	Number of plants per genotype	at least 20 plants

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² resistentie@naktuinbouw.nl

³ resistencias@inia.es

⁴ Harmores 2 CPVO project (<http://www.cpvo.europa.eu/main/en/home/documents-and-publications/technical-projects-reports>)

9.2	Number of replicates	-
9.3	Control varieties	<p><u>TMV: 0:</u></p> <ul style="list-style-type: none"> - Susceptible controls: Lamu, Pepita, Piquillo - Resistant controls: Fehérözön, Yolo Wonder <p><u>PMMoV: 1.2:</u></p> <ul style="list-style-type: none"> - Susceptible controls: Fehérözön, Lamu, Yolo Wonder - Resistant controls: Ferrari, Novi 3 <p><u>PMMoV: 1.2.3:</u></p> <ul style="list-style-type: none"> - Susceptible controls: Ferrari, Yolo Wonder - Resistant controls: Friendly, Tom 4 <p>For PMMoV: 1.2.3, it is advised to choose Ferrari as susceptible control because it is resistant to PMMoV: 1.2 or to add the differentials in tests to confirm the group.</p>
9.4	Test design	add non-inoculated plants
9.5	Test facility	Climate room or greenhouse
9.6	Temperature	20-25°C
9.7	Light	12 hours or longer
9.8	Season	-
9.9	Special measures	-
10.	Inoculation	
10.1	Preparation inoculum	1 g leaf with symptoms with 10 mL PBS or similar buffer or dilution of juice in water. Homogenize, add carborundum to buffer
10.2	Quantification inoculum	-
10.3	Plant stage at inoculation	<u>TMV: 0</u> , cotyledons to first leaf stage <u>PMMoV: 1.2 and PMMoV: 1.2.3</u> , cotyledon stage
10.4	Inoculation method	rubbing with the virus suspension
10.5	First observation	<p><u>TMV:0:</u></p> <p>4-7 days post-inoculation for observation of local necrosis.</p> <p><u>PMMoV: 1.2 and PMMoV: 1.2.3:</u></p> <p>4-7 days post-inoculation for observation of local necrotic lesions which can lead to cotyledon drop. After this date these necrosis can hardly be seen on fallen cotyledons</p>
10.6	Second observation	<p><u>TMV: 0:</u></p> <p>two weeks post-inoculation for observation of symptoms of susceptibility.</p> <p><u>PMMoV: 1.2 and PMMoV: 1.2.3:</u></p> <p>two weeks post-inoculation for observation of symptoms of susceptibility.</p>
10.7	Final observations	<p><u>TMV: 0:</u></p> <p>three weeks post-inoculation.</p> <p><u>PMMoV: 1.2 and PMMoV: 1.2.3:</u></p> <p>three weeks post-inoculation.</p> <p>For TMV:0, PMMoV: 1.2 and PMMoV: 1.2.3, two of these three observations may be sufficient; the third notation is optional for observation of evolution of symptoms (depending on symptoms on controls or heterogeneous behaviour)</p>
11.	Observations	
11.1	Method	Visual
11.2	Observation scale	<p><u>TMV: 0:</u></p> <ul style="list-style-type: none"> - Susceptibility: mosaic (Aucuba in case of Aucuba strain as Vi-6), growth reduction, death of plants. - Resistance: local necrotic lesions which can lead to leave drop, systemic necrosis, vein necrosis, stem necrosis. <p><u>PMMoV: 1.2 and PMMoV: 1.2.3:</u></p> <ul style="list-style-type: none"> - Susceptibility: mosaic (green), growth reduction. - Resistance: local necrotic lesions which can lead to cotyledon drop, systemic necrosis
11.3	Validation of test	Evaluation of variety resistance should be calibrated with results of resistant and susceptible controls

11.4	Off-types	-
12.	Interpretation of data in terms of UPOV characteristic states	absent [1] susceptible, see 11.2 present [9] resistant, see 11.2
13.	Critical control points	- For TMV: 0, plants with no symptoms at all have to be interpreted as escapes of inoculation. - Recommended dates of notation should be adapted depending of expression of symptoms on controls. - Environmental conditions can have an effect on the expression of symptoms over time. In this case a third notation could be necessary.

Ad. 55: Resistance to Tobamovirus - *Pepper mild mottle virus* - Group 2 (PMMoV: 1.2)

See Ad. 54

Ad. 56: Resistance to Tobamovirus - *Pepper mild mottle virus* - Group 3 (PMMoV: 1.2.3)

See Ad. 54

Ad. 57: Resistance to *Potato Y virus* (PVY) - Pathotype 0 (PVY: 0)

1.	Pathogen	<i>Potato Y virus</i> (PVY)
2.	Quarantine status	No
3.	Host species	Sweet pepper, hot pepper, paprika and chili – <i>Capsicum annuum</i> L
4.	Source of inoculum	GEVES ⁵ (FR), Naktuinbouw ⁶ (NL) or INIA - CSIC ⁷ (SP)
5.	Isolate	- For PVY: 0 strain zb6 (the test protocol has been validated in a CPVO co-funded project ⁸ with this isolate/race). - PVY race 1 - PVY race 2
6.	Establishment isolate identity	genetically defined pepper controls (ref. ISF site: nov. 2020: Differential Hosts – International Seed Federation (worldseed.org))

Differential Host	gene present	PVY: 0	PVY: 1	PVY: 1.2
Early Cal Wonder, Yolo Wonder	<i>pvr 0</i>	S	S	S
PI152225	<i>pvr 1</i>	HR	HR	-
Yolo Y	<i>pvr1¹ (pvr 2¹)</i>	HR	S	S
Florida VR2	<i>pvr1² (pvr 2²)</i>	HR	HR	S
Florida VR4, Del Rey Bell, Agronomico 10	<i>pvr3</i>	HR	HR	HR
Serrano Criollo de Morelos 334	<i>pvr4</i>	HR	HR	HR

S= susceptible; HR= highly resistant

Note: In some scientific publications *pvr 2¹* is referred to as *pvr 1¹*. Similarly, *pvr 2²* is referred to as *pvr 1²*

7.	Establishment pathogenicity	Test on susceptible plants
8.	Multiplication inoculum	
8.1	Multiplication medium	Regeneration of the virus on plant material before inoculum preparation
8.2	Multiplication variety	On susceptible pepper variety, PVY races may be multiplied on varieties which are selective for each particular race. For PVY: 0, because tobacco <i>Nicotiana tabacum</i> cv. <i>Xanthi-nc</i> has large leaves and can produce a lot of inoculum and has a faster multiplication, it is recommended for the multiplication.

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⁸ Harmores 2 CPVO project (<http://www.cpvo.europa.eu/main/en/home/documents-and-publications/technical-projects-reports>)

8.3	Plant stage at inoculation	see 10.3
8.4	Inoculation medium	see 10.1
8.5	Inoculation method	see 10.4
8.6	Harvest of inoculum	Symptomatic fresh leaves
8.7	Check of harvested inoculum	Option: on <i>Nicotiana tabacum</i> cv. <i>Xanthi-nc</i> , check mosaic presence and local lesion absence (contamination by Tobamovirus) after 5-7 days.
8.8	Shelf life/viability inoculum	fresh > 1 day, desiccated > 1 year. Because problem of stability of PVY: 0, shipments are recommended to be done with fresh infected leaves
9.	Format of the test	
9.1	Number of plants per genotype	at least 20 plants
9.2	Number of replicates	-
9.3	Control varieties	<u>PVY: 0:</u> - Susceptible controls: Ferrari, Piquillo, Yolo Wonder - Resistant controls: Andalus, Vidi, Yolo Y <u>PVY: 1:</u> - Susceptible controls: Yolo Wonder, Yolo Y - Resistant controls: Florida VR2 <u>PVY: 1.2:</u> - Susceptible controls: Florida VR2, Yolo Wonder, Yolo Y - Resistant controls: Serrano Criollo de Morelos
9.4	Test design	add non inoculated plants
9.5	Test facility	Climate room or greenhouse. In case of test in greenhouse during period of low daylight, shadowy area should not be used
9.6	Temperature	18-25°C
9.7	Light	12 hours or longer
9.8	Season	-
9.9	Special measures	For PVY: 0, it is advised to choose Yolo Y as resistant control or to add the differentials in tests to be able to observe a possible contamination by PVY: 1 or 1.2
10.	Inoculation	
10.1	Preparation inoculum	1 g leaf with symptoms with 4 mL PBS with carborundum (80mg) and activated carbon (80mg) or similar buffer, homogenize
10.2	Quantification inoculum	-
10.3	Plant stage at inoculation	PVY: 0: cotyledons stage PVY: 1 and 1.2: cotyledons stage or first pointing leaf stage
10.4	Inoculation method	rubbing with the virus suspension
10.5	Final observations	Three weeks post-inoculation
11.	Observations	
11.1	Method	Visual
11.2	Observation scale	<u>Susceptibility:</u> mosaic (can be very light/faint), growth reduction, vein banding and vein necrosis. <u>Resistance:</u> no symptoms
11.3	Validation of test	Evaluation of variety resistance should be calibrated with results of resistant and susceptible controls.
11.4	Off-types	-
12.	Interpretation of data in terms of UPOV characteristic states	absent [1] susceptible, see 11.2 present [9] resistant, see 11.2
13.	Critical control points	Recommended dates of notation should be adapted depending of expression of symptoms on controls.

Ad. 58: Resistance to *Potato Y virus* (PVY) - Pathotype 1 (PVY: 1)

See Ad. 57

Ad. 59: Resistance to *Potato Y virus* (PVY) - Pathotype 1.2 (PVY: 1.2)

See Ad. 57

Ad. 60: Resistance to *Phytophthora capsici* (Pc)

1.	Pathogen	<i>Phytophthora capsici</i> (Pc)
2.	Quarantine status	no
3.	Host species	<i>Capsicum annuum</i>
4.	Source of inoculum	INRAE GAFL (FR)
5.	Isolate	moderately aggressive (e.g. strain P0277)
6.	Establishment isolate identity	on standards Jupiter, Yolo Wonder (susceptible), Favolor (moderately resistant), Solario, Phyto 636 (resistant)
7.	Establishment pathogenicity	in biotest on plants
8.	Multiplication inoculum	
8.1	Multiplication medium	V8 juice-agar (1%) or 10% V8A or PDA+
8.2	Multiplication variety	-
8.3	Plant stage at inoculation	-
8.4	Inoculation medium	10% V8A or PDA+
8.5	Inoculation method	see 10.4
8.6	Harvest of inoculum	-
8.7	Check of harvested inoculum	-
8.8	Shelf life/viability inoculum	10% V8A 3 months, PDA+ 2 months
9.	Format of the test	
9.1	Number of plants per genotype	at least 20 plants (2 untreated plants)
9.2	Number of replicates	e.g. 1
9.3	Control varieties	Jupiter, Yolo Wonder (susceptible), Favolor (moderately resistant), Solario (resistant)
9.4	Test design	-
9.5	Test facility	glasshouse
9.6	Temperature	22°C d/n
9.7	Light	at least 12h
9.8	Season	-
9.9	Special measures	-
10.	Inoculation	
10.1	Preparation inoculum	growing on Petri dishes
10.2	Quantification inoculum	-
10.3	Plant stage at inoculation	first flower bud
10.4	Inoculation method	Stem is cut just below point of first branching, a 4mm-agar plug is placed carefully on the wound and covered with aluminum foil
10.5	First observation	7 days post inoculation
10.6	Second observation	14 days post inoculation
10.7	Final observations	21 days post inoculation
11.	Observations	
11.1	Method	visual, comparative or measurement of stem necrosis length; for repeated measurements, the stem is marked with permanent ink
11.2	Observation scale	
	- susceptible	e.g. length increase > 0.8 cm/week
	- moderately resistant	e.g. length increase ≥ 0.5 cm ≤ 0.8 cm/week
	- highly resistant	e.g. length increase < 0.5 cm/week
11.3	Validation of test	Evaluation of variety resistance should be based on the stem necrosis increase compared to the control varieties.
11.4	Off-types	maximum 1 on 20 plants
12.	Interpretation of data in terms of UPOV characteristic states	Absent..... [1] susceptible Present..... [9] moderately resistant and highly resistant
13.	Critical control points	- Absence of differential interactions between host and pathogen - Maintenance of viability of the strains in the collection

Ad. 61: Resistance to *Cucumber mosaic virus* (CMV)

1.	Pathogen	<i>Cucumber mosaic virus</i> (CMV)
2.	Quarantine status	no
3.	Host species	<i>Capsicum annuum</i>
4.	Source of inoculum	INRAE GAFL (FR)
5.	Isolate	e.g. 'Fulton'
6.	Establishment isolate identity	-
7.	Establishment pathogenicity	-
8.	Multiplication inoculum	
8.1	Multiplication medium	living plant
8.2	Multiplication variety	e.g. <i>Vinca rosea</i>
8.3	Plant stage at inoculation	-
8.4	Inoculation medium	0.03 M PBS + 0.1% DIECA
8.5	Inoculation method	rubbing with carborundum
8.6	Harvest of inoculum	1 g on 4 ml buffer
8.7	Check of harvested inoculum	-
8.8	Shelf life/viability inoculum	-
9.	Format of the test	
9.1	Number of plants per genotype	at least 50 plants
9.2	Number of replicates	e.g. 1
9.3	Control varieties	Yolo Wonder (susceptible), Ducato (moderately resistant), Alby, Favolor (resistant)
9.4	Test design	-
9.5	Test facility	-
9.6	Temperature	20-22°C
9.7	Light	12h
9.8	Season	-
9.9	Special measures	-
10.	Inoculation	
10.1	Preparation inoculum	-
10.2	Quantification inoculum	-
10.3	Plant stage at inoculation	cotyledon, before emergence of first leaf (12-13 days after sowing)
10.4	Inoculation method	rubbing cotyledons with carborundum, followed by 48h darkness
10.5	First observation	10 days post inoculation
10.6	Second observation	15 days post inoculation
10.7	Final observations	21 days post inoculation
11.	Observations	
11.1	Method	visual, comparative
11.2	Observation scale	
	- susceptible	many local lesions, mosaic
	- moderately resistant	intermediate symptoms
	- highly resistant	few local lesions, no or light symptoms
11.3	Validation of test	Evaluation of variety resistance should be calibrated with results of resistant and susceptible controls.
11.4	Off-types	maximum 1 on 20 plants
12.	Interpretation of data in terms of UPOV characteristic states	Absent..... [1] susceptible Present.....[9] moderately resistant and highly resistant
13.	Critical control points	-

Ad. 62: Resistance to *Tomato spotted wilt virus* Pathotype 0 (TSWV: 0)

1.	Pathogen	<i>Tomato spotted wilt virus</i> , Pathotype 0 (TSWV: 0)
2.	Quarantine status	yes
3.	Host species	<i>Capsicum annuum</i>
4.	Source of inoculum	GEVES (FR), Naktuinbouw (NL), INIA CSIC (ES)
5.	Isolate	e.g. LYE 51 or Br-01
6.	Establishment isolate identity	-
7.	Establishment pathogenicity	Test on susceptible plant or <i>Nicotiana benthamiana</i> , <i>N. rustica</i>
8.	Multiplication inoculum	
8.1	Multiplication medium	living plant
8.2	Multiplication variety	Yolo Wonder or <i>N. benthamiana</i> , <i>N. rustica</i>
8.3	Plant stage at inoculation	Cotyledons fully developed or at "first leaf" pointed stage or 1- 3 leaves
8.4	Inoculation medium	Ice-cold buffer suspension or 0.03 M PBS + optional addition of 0.1% sodium sulfite freshly added
8.5	Inoculation method	Rubbing with carborundum
8.6	Harvest of inoculum	-
8.7	Check of harvested inoculum	-
8.8	Shelf life/viability inoculum	Stability in ice cold suspension ca. 15-20 minutes
9.	Format of the test	
9.1	Number of plants per genotype	at least 20 plants
9.2	Number of replicates	e.g. 1
9.3	Control varieties	Lamuyo, Yolo Wonder (susceptible), Galileo, Jackal, Jackpot, Prior (resistant)
9.4	Test design	-
9.5	Test facility	Growth chamber or insect proof glasshouse
9.6	Temperature	18-20°C or 20-22°C
9.7	Light	12h
9.8	Season	All seasons, but winter reduces the risk of thrips infestation
9.9	Special measures	Biohazard sign on compartment for countries with a TSWV quarantine status
10.	Inoculation	
10.1	Preparation inoculum	-
10.2	Quantification inoculum	-
10.3	Plant stage at inoculation	Cotyledons fully developed /at "first leaf" pointed stage or 1-3 leaves
10.4	Inoculation method	Rubbing with carborundum, then apply shading or darkness for 24h Option: repeat the inoculation 2-3 days later to reduce accidental escapes
10.5	First observation	5-6 days to 10 - 15 days post inoculation
10.6	Second observation	10-11 days post inoculation to 15 - 21 days post inoculation
10.7	Final observations	21 days post inoculation
11.	Observations	
11.1	Method	Visual, comparative
11.2	Observation scale	
		Susceptibility: mosaic on young leaf, some leaf malformation Resistance: necrosis or only mechanical damage
11.3	Validation of test	Evaluation of variety resistance should be calibrated with results of resistant and susceptible controls.
11.4	Off-types	maximum 1 on 20 plants
12.	Interpretation of data in terms of UPOV characteristic states	absent [1] susceptible, see 11.2 present [9] resistant, see 11.2
13.	Critical control points	- Monitor and control the presence of thrips. TSWV is transmitted by thrips (<i>Thrips tabaci</i> and <i>Frankliniella occidentalis</i>). TSWV has a broad host range. - After a few multiplications the virus could be ineffective. New isolates can be obtained from practice by harvesting fruits of L4 pepper varieties infected naturally with TSWV. The fruits are kept at -70°C temperature. The presence of other viruses must be checked before using this material.

Ad. 63: Resistance to *Xanthomonas* spp (ex *Xanthomonas campestris* pv. *vesicatoria*) (X spp (ex Xcv)) - Pathotype 1

1.	Pathogen	<i>Xanthomonas</i> spp (ex <i>Xanthomonas campestris</i> pv. I) (X spp (ex Xcv))
2.	Quarantine status	-
3.	Host species	<i>Capsicum annuum</i>
4.	Source of inoculum	Natural; to be taken from any source of infection in the field
5.	Isolate	Expected reactions on resistant standard varieties
6.	Establishment isolate identity	on differentials

Differential	Pathotype 1	Pathotype 2	Pathotype 3
Early California Wonder	S	S	S
Early California Wonder-10R (gene Bs1)	S	R	S
Early California Wonder-20R (gene Bs2)	R	R	R
Early California Wonder-30R (gene Bs3)	R	S	S
PI 235047 (gene Bs4)	R	S	R

7.	Establishment pathogenicity	-
8.	Multiplication inoculum	-
8.1	Multiplication medium	A bacterial growth medium, e.g. LPGA
8.2	Multiplication variety	-
8.3	Plant stage at inoculation	-
8.4	Inoculation medium	-
8.5	Inoculation method	-
8.6	Harvest of inoculum	48h culture
8.7	Check of harvested inoculum	-
8.8	Shelflife/viability inoculum	-
9.	Format of the test	-
9.1	Number of plants per genotype	at least 20 plants
9.2	Number of replicates	e.g. 1
9.3	Control varieties	Fehérözön, Yolo Wonder (susceptible), Emiro, Filidor, Gotico, San Marco, Solanor (resistant)
9.4	Test design	-
9.5	Test facility	-
9.6	Temperature	20-26°C day/night
9.7	Light	30.000 lux suggested, 16h/day
9.8	Season	-
9.9	Special measures	80% RH
10.	Inoculation	-
10.1	Preparation inoculum	Harvest cells from LPGA plate after 48 h growing
10.2	Quantification inoculum	10 ⁷ -10 ⁸ cells per ml (Stronger reaction with the higher concentration.)
10.3	Plant stage at inoculation	6-8 true leaves
10.4	Inoculation method	Infiltration into abaxial surface of the interveinal region on either side of the midrib of a fully expanded leaf in 13-20mm diameter spots
10.5	First observation	2-5 days post inoculation
10.6	Second observation	6-8 days post inoculation
10.7	Final observations	10-14 days post inoculation
11.	Observations	-
11.1	Method	Visual, comparative
11.2	Observation scale	Susceptibility: Water soaking near infiltration site Resistance: Necrotic reaction at infiltration site
11.3	Validation of test	Evaluation of variety resistance should be calibrated with results of resistant and susceptible controls.
11.4	Off-types	maximum 1 on 20 plants

12.	Interpretation of data in terms of UPOV characteristic states	absent present	[1] [9]	susceptible, see 11.2 resistant, see 11.2
13.	Critical control points	-		

Ad. 64: Resistance to *Xanthomonas* spp (ex *Xanthomonas campestris* pv. *vesicatoria*) (X spp (ex Xcv)) - Pathotype 2

See Ad. 63

Ad. 65: Resistance to *Xanthomonas* spp (ex *Xanthomonas campestris* pv. *vesicatoria*) (X spp (ex Xcv)) - Pathotype 3

See Ad. 63

Ad. 66: Resistance to *Meloidogyne incognita* (Mi)

1.	Pathogen	<i>Meloidogyne incognita</i> (Mi)
2.	Quarantine status	-
3.	Host species	Sweet pepper, hot pepper, paprika and chili – <i>Capsicum annum</i> L.
4.	Source of inoculum	GEVES ⁹ (F)
5.	Isolate	non-resistance breaking
6.	Establishment isolate identity	use pepper standards
7.	Establishment pathogenicity	use pepper standards
8.	Multiplication inoculum	
8.1	Multiplication medium	living plant of pepper or tomato
8.2	Multiplication variety	susceptible variety
8.3	Plant stage at inoculation	2 leaves stage
8.5	Inoculation method	Deposit of piece of contaminated roots in soil (around 5-10g per plant, to adapt depending on the population aggressivity)
8.6	Harvest of inoculum	6 to 10 weeks after inoculation, root systems are cut with scissors into pieces of about 1 cm length
8.7	Check of harvested inoculum	visual check for presence of root knots and ripe egg masses
8.8	Shelflife/viability inoculum	1 day
9.	Format of the test	
9.1	Number of plants per genotype	at least 30 plants, plus at least 10 non-inoculated plants to observe if a possible lack of germination is due to nematode or not. It is recommended to sow more seeds to be sure to get enough plants.
9.2	Number of replicates	At least 2, preferably 3.
9.3	Control varieties	Susceptible: Tom 4 and Yolo Wonder (as additional susceptible control for reduced susceptibility, indicating the border between S and R) Resistant: Capital and W4
9.4	Test design	3 replicates of 10 plants per variety, in separate trays with contaminated substrate (70% soil +30% sand) to allow statistical analysis. 10 plants in a separate tray with NON contaminated substrate.
9.5	Test facility	greenhouse or climate room
9.6	Temperature	20-26°C, the temperature must be adapted depending on the aggressivity of the test to obtain expected response of controls but should not be above 26°C.
9.7	Light	at least 12 h per day
10.1	Preparation inoculum	Small pieces of diseased roots mixed with soil
10.2	Quantification inoculum	The ratio is depending on aggressiveness of test and laboratories conditions (e.g. between 15g to 30g of infested roots, for 40 plants in a tray of 30*30 cm containing approximately 3.5 kg of substrate,), galls should be mixed homogeneously with the soil.
10.3	Plant stage at inoculation	seed
10.4	Inoculation method	Seeds sown in soil contaminated with infested roots homogeneously mixed with soil

⁹ GEVES; matref@geves.fr

10.5	First observation	-
10.6	Second observation	-
10.7	Final observations	Around 45 days after inoculation depending on test conditions (temperature, season)
11.	Observations	
11.1	Method	root inspection
11.2	Observation scale	<p>Class 0: healthy plant, no galls</p> <p>Class 1: few and little galls which are difficult to find (for example less than 5)</p> <p>Class 2: few galls, easy to observe but on few roots, still a lot of roots without galls, no chains</p> <p>Class 3: many individual galls on most but not all roots, presence of chains</p> <p>Class 4: many galls on all roots, can lead to dead plants and may suppress emergence.</p>
11.3	Validation of test	Evaluation of variety resistance should be calibrated with results of resistant and susceptible controls.
11.4	Off-types	resistant varieties may have a few plants with a few galls
12.	Interpretation of data in terms of UPOV characteristic states	<p>Variety very similar to resistant control is judged as resistant:</p> <p>Variety very similar to susceptible controls is judged as susceptible: Resistance is absent (1);</p> <p>If significantly different from resistant and susceptible controls (notations are between resistant and susceptible controls), the variety is judged as resistant; Yolo Wonder is the border control variety for susceptibility. Varieties with higher resistance than Yolo wonder are judged as resistant: Resistance is present (9);</p> <p>If results are not clear, statistical analysis is advised.</p> <p><i>The analysis of raw data of the couple Mi / Pepper is planned in the Pathostat tool (free statistic analysis dedicated to quantitative disease resistances) https://pathostat.geves.fr</i></p>
13.	Critical control points	<p>Avoid rotting of roots; high temperature causes breakdown of resistance. In case of an aggressive test, put seeds in a layer of non-contaminated soil or decrease the quantity of inoculum.</p> <p>In class 4 heavy gall development is seldom observed, normally it can occur as loss of seedlings.</p> <p>If germination of non-inoculated seeds is 100%, non-germinated inoculated seeds are expected to be in class 4. If germination of non-inoculated seeds is less than 100%, equal lower germination percentage can be expected concerning the inoculated seeds.</p>

9. LITERATURE

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10. TECHNICAL QUESTIONNAIRE

The Technical Questionnaire is available on the [CPVO website](#) under the following reference:
CPVO/TQ-076/3 – *Capsicum annuum* L. – sweet pepper, hot pepper, paprika, chili