



## **PROTOCOL FOR TESTS ON DISTINCTNESS, UNIFORMITY AND STABILITY**

***Cucurbita pepo* L.**

**VEGETABLE MARROW, SQUASH**

UPOV Code: CUCUR\_PEP

**Adopted on 31/03/2025**

**Entry into force on 01/04/2025**

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## **1. SUBJECT OF THE PROTOCOL AND REPORTING**

### **1.1 Scope of the technical protocol**

This Technical Protocol applies to all varieties of *Cucurbita pepo* 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](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/119/4 Rev dated 09/08/2024 (<https://www.upov.int/edocs/tgdocs/en/tg119.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**

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

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

The testing of a variety may be concluded when the entrusted examination office 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](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**

For 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.

#### **3.6.2 Living Plant Material**

For seed propagated varieties, the EO shall collect and maintain living plant material of varieties of the species concerned in the variety collection.

#### **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**

For 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.

## **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](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](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. 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.

### **(a) Cross-pollinated varieties**

The assessment of uniformity should be according to the recommendations for cross-pollinated varieties in the UPOV-General Introduction to DUS.

### **(b) Hybrid varieties**

For the assessment of uniformity of hybrid varieties, a population standard of 1% and an acceptance probability of at least 95% should be applied to off-types excluding clearly recognisable inbred plants. In addition, a population standard of 1% with the same acceptance probability should be applied to clearly recognisable inbred plants. In the case of a sample size of 20 plants, 1 off-type is allowed.

### **4.3 Stability**

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](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 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) Fruit: type (case outlined in page 8 of the protocol)
  - b) Plant: growth habit (characteristic 4)
  - c) Plant: branching (characteristic 5)
  - d) Leaf blade: incisions (characteristic 13)
  - e) Leaf blade: silvery patches (characteristic 15)
  - f) Leaf blade: relative area covered by silvery patches (characteristic 16)
  - g) Fruit: general shape (characteristic 30)
  - h) Fruit: main colour of skin (characteristic 50)
- 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".

b) types of varieties of *Cucurbita pepo* L.

<b>Fruit: type</b>	<b>Fruit: general shape (Ch. 30)</b>	<b>Plant: growth habit (Ch.4)</b>		<b>Fruit: grooves (Ch. 46)</b>	<b>Fruit: ribs (Ch.48)</b>	<b>Fruit: warts on skin (Ch.62)</b>	<b>Ripe fruit: main colour of skin (Ch.69)</b>	<b>Ripe fruit: lignified rind (Ch.75)</b>	<b>Ripe fruit: colour of flesh (Ch.74)</b>	<b>Ripe fruit: structure of flesh (Ch.76)</b>	<b>Example varieties</b>
Pumpkin	From flattened globular to elliptical globular			present	absent	absent		absent	orange		Halloween, Little Boo, Small Sugar
Miniature Pumpkin	Transverse elliptical	trailing	very small	present	absent	absent		absent	orange		Jack Be Little
Scallop	Flattened disc shaped with equatorial margin	bush or semi-trailing		absent	absent			present	cream		Patty Pan, Scallopini
Acorn	Top shaped with furrows	bush						absent	orange		Table Queen
Neck	Bottle shaped with pointed blossom end	bush				present	Orange	present			Early Prolific Straightneck, Early Summer Crookneck
Zucchini	From pear-shaped to elliptical to cylindrical to club-shaped							present	cream		Ambassador, Beirut, Clarita, Elite, Ibis, Romano
Rounded Zucchini	Globular	bush				absent		present	cream		De Nice à fruit rond, Redondo
Delicata	Elliptical	trailing		present	absent	absent					Delicata
Spaghetti Squash	Elliptical	trailing				absent		absent	dark yellow to orange	fibrous	Pasta, Vegetable Spaghetti
Rondini	Globular	trailing	very small		absent	absent		present	yellow		Little Gem
Ölkürbis	Globular	trailing			present	absent					Markant
Other											



## 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 8.1
(*)	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
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## 7. TABLE OF CHARACTERISTICS

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note
1.	1.		Seedling: shape of cotyledons		
			narrow elliptic	Bianchini	1
			elliptic	Cora, Tivoli	2
			broad elliptic	Cinderella, Goldi	3
			circular	Early Summer Crookneck	4
			obovate		5
2.	2.		Seedling: intensity of green colour of cotyledons		
			very light	Sunburst	1
			very light to light		2
			light	Bianchini	3
			light to medium		4
			medium	Cora	5
			medium to dark		6
			dark	Lidia	7
			dark to very dark		8
			very dark	Saray	9
3.	3. (*)		Seedling: cross section of cotyledons		
			concave		1
			straight	Sunburst	2
			convex	Bianchini, Early Summer Crookneck	3
4.	4. (*)		Plant: growth habit		
			bush	Greyzini	1
			semi-trailing	Cinderella, Everest, Twickers	2
			G	trailing	Becky, Long Green Trailing

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note
5.	5. (*)		<b>Plant: branching</b>		
			absent	Goldi	1
G			present	Patty Green Tint	9
6.	6.		<b>Plant: degree of branching</b>		
			very weak	Cora	1
			very weak to weak		2
			weak	Karioka, Verdi	3
			weak to medium		4
			medium	Gold Rush, Tivoli	5
			medium to strong		6
			strong	Blanche non coureuse 3, Jack O'Lantern, Early Summer Crookneck	7
			strong to very strong		8
			very strong	Patty Green Tint, Sweet Dumpling	9
7.	7.		<b>Bush varieties only: Plant: attitude of petiole (excluding lower external leaves)</b>		
			erect	Blanche non coureuse 3, Early Summer Crookneck	1
			erect to semi-erect	Sardane	2
			semi-erect	Arlesa	3
			semi-erect to horizontal	Goldi	4
			horizontal	Ambassador	5
8.	8.		<b>Stem: colour</b>		
			completely green	Becky	1
			partly green and partly yellow	Autumn Gold	2

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note
9.	9.		<b>Stem: intensity of green colour</b>		
			very light	Maayan	1
			very light to light		2
			light	Bianchini	3
			light to medium		4
			medium	Cinderella	5
			medium to dark		6
			dark	Greyzini	7
			dark to very dark		8
			very dark	Goldrush	9
10.	10.		<b>Stem: mottling</b>		
			absent	Cinderella	1
			present	Cora	9
11.	11.		<b>Stem: tendrils</b>		
			absent to rudimentary	Goldrush, Sylvana	1
			well developed	Baby Bear, Greyzini	2
12.	12.		<b>Leaf blade: size</b>		
			very small	Becky	1
			very small to small		2
			small	Small Sugar	3
			small to medium		4
			medium	Ambassador	5
			medium to large		6
			large	Kriti	7
			large to very large		8
			very large		9

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note
<b>13. (+)</b>	<b>13. (*)</b>		<b>Leaf blade: incisions</b>		
			absent or very shallow	Scallopini	1
			very shallow to shallow		2
			shallow	Everest	3
			shallow to medium		4
			medium	Jackpot	5
			medium to deep		6
			deep	Civac	7
			deep to very deep		8
<b>G</b>			very deep	Isotta	9
<b>14.</b>	<b>14.</b>		<b>Leaf blade: intensity of green colour of upper surface</b>		
			very light		1
			very light to light		2
			light	Ghazzi	3
			light to medium		4
			medium	Cora	5
			medium to dark		6
			dark	Everest	7
			dark to very dark		8
			very dark		9
<b>15.</b>	<b>15. (*)</b>		<b>Leaf blade: silvery patches</b>		
			absent	Black Forest, Scallopini	1
<b>G</b>			present	Civac	9

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note
16.	16.		<b>Leaf blade: relative area covered by silvery patches</b>		
			very small	Albo	1
			very small to small		2
			small	Aziz	3
			small to medium		4
			medium	Ambassador	5
			medium to large		6
			large	Cora	7
			large to very large		8
<b>G</b>			very large	Summerstar	9
17.	17.		<b>Petiole: length</b>		
			very short		1
			very short to short		2
			short	Jack be Little, Karioka	3
			short to medium		4
			medium	Goldi	5
			medium to long		6
			long	Autumn Gold, Baikal	7
			long to very long		8
			very long		9

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note
<b>18.</b>	<b>18.</b>		<b>Petiole: number of prickles</b>		
			absent or very few	Kojac	1
			very few to few		2
			few	Opaline	3
			few to medium		4
			medium	Spidy	5
			medium to many		6
			many	White Bush Scallop	7
			many to very many		8
			very many	Early Summer Crookneck	9
<b>19.</b>	<b>19. (*)</b>		<b>Female flower: ring at inner side of corolla</b>		
			absent	Cinderella, Greyzini	1
			present	Aurore	9
<b>20.</b>	<b>20.</b>		<b>Female flower: colour of ring at inner side of corolla</b>		
			yellow	Diamant, Patro	1
			green	Aurore, Early White Bush Scallop, President	2
			yellow and green	Pueblo	3

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note
21.	21.		<b><u>Only varieties with green ring at inner side of corolla: Female flower: intensity of colour of green ring at inner side of corolla</u></b>		
			very weak		1
			very weak to weak		2
			weak	Maya, Sardane	3
			weak to medium		4
			medium	Samba, Senator	5
			medium to strong		6
			strong	Aristocrat, Diamant	7
			strong to very strong		8
			very strong		9
22.	22.		<b>Male flower: ring at inner side of corolla</b>		
			absent	Bianchini	1
			present	Goldi	9
23.	23. (*)		<b>Male flower: colour of ring at inner side of corolla</b>		
			yellow	Afrodite, Patro, Zyzo	1
			green	Austral, Belor, Goldi	2
			yellow and green	Alice, Carmina, Green Gem, Ibis	3



CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note
24.	24.		<b><u>Only varieties with green ring at inner side of corolla: Male flower: intensity of green colour of ring at inner side of corolla</u></b>		
			very weak		1
			very weak to weak		2
			weak	Cora	3
			weak to medium		4
			medium	Verdi	5
			medium to strong		6
			strong	Goldi	7
			strong to very strong		8
			very strong		9
25.	25.		<b><u>Only Zucchini type varieties: Young fruit: ratio length/maximum diameter</u></b>		
			very small	Ghazzi	1
			very small to small		2
			small	Opal	3
			small to medium		4
			medium	Cora	5
			medium to large		6
			large	Carlotta	7
			large to very large		8
			very large	Spidy	9

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note
26. (+)	26.		<b><u>Only Zucchini and Rounded Zucchini type varieties:</u></b> Young fruit: general shape		
			globular	De Nice à fruit rond	1
			pear shaped	Clarita	2
			tapered elliptical	Top Kapi	3
			elliptical	Table Dainty	4
			cylindrical	Ambassador, Ibis	5
			tapered cylindrical		6
27.	27.		<b>Young fruit: main colour of skin (excluding colour of ribs or grooves)</b>		
			white	White Bush Scallop	1
			cream	Tivoli	2
			yellow	Goldi	3
			green	Elite, Opal, Romano	4
			partly white and partly yellow		5
			partly white and partly green		6
28.	28.		<b><u>Only varieties with yellow colour of skin:</u></b> Young fruit: intensity of yellow colour of skin (as for 27)		
			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
29.	29.		<b><u>Only varieties with green colour of skin: Young fruit: intensity of green colour of skin (as for 27)</u></b>		
			very light	Clarita, Goya, Patty Green Tint	1
			very light to light		2
			light	Arlika	3
			light to medium		4
			medium	Baccara	5
			medium to dark		6
			dark	Arlesa, Sandra, Zefira	7
			dark to very dark		8
			very dark	Carnaval, Corsair	9
30. (+)	30. (*)		<b>Fruit: general shape</b>		
			disc shaped		1
			transverse elliptical		2
			transverse broad elliptical		3
			globular		4
			top shaped		5
			broad elliptical		6
			ovate		7
			elliptical		8
			cylindrical		9
			pear shaped		10
			bottle shaped		11
<b>G</b>			club shaped		12

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note
31.1	31.1 (*)		<b><u>Only Scallop type varieties:</u> Fruit: length</b>		
			very short		1
			very short to short		2
			short	Bennings Green Tint	3
			short to medium		4
			medium	Sunburst	5
			medium to long		6
			long	Yellow Bush Scallop	7
			long to very long		8
			very long		9
31.2	31.2 (*)		<b><u>Only Acorn type varieties:</u> Fruit: length</b>		
			very short		1
			very short to short		2
			short	Table Gold	3
			short to medium		4
			medium	Swan White Acorn	5
			medium to long		6
			long	Ebony Table Queen	7
			long to very long		8
			very long		9

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note
31.3	31.3 (*)		<b><u>Only Neck type varieties:</u> Fruit: length</b>		
			very short		1
			very short to short		2
			short	Wryneck	3
			short to medium		4
			medium	Early Summer Crookneck	5
			medium to long		6
			long	Sunbar	7
			long to very long		8
			very long		9
31.4	31.4 (*)		<b><u>Only Zucchini type varieties:</u> Fruit: length</b>		
			very short	Jericho	1
			very short to short		2
			short	Jedida	3
			short to medium		4
			medium	Cora	5
			medium to long		6
			long	Carlotta	7
			long to very long		8
			very long	Altea	9

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note
32.1	32.1		<b><u>Only Scallop type varieties:</u> Fruit: maximum diameter</b>		
			very small		1
			very small to small		2
			small	Scallopini	3
			small to medium		4
			medium	Yellow Bush Scallop	5
			medium to large		6
			large	White Bush Scallop	7
			large to very large		8
			very large		9
32.2	32.2		<b><u>Only Acorn type varieties:</u> Fruit: maximum diameter</b>		
			very small		1
			very small to small		2
			small	Table Gold	3
			small to medium		4
			medium	Table King	5
			medium to large		6
			large	Swan White Acorn	7
			large to very large		8
			very large		9

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note
32.3	32.3		<b><u>Only Zucchini type varieties:</u> Fruit: maximum diameter</b>		
			very small		1
			very small to small		2
			small	Goldi	3
			small to medium		4
			medium	Opal	5
			medium to large		6
			large	Jericho, Spidy	7
			large to very large		8
			very large		9
33.1	33.1		<b><u>Only Scallop type varieties:</u> Fruit: ratio length/maximum diameter</b>		
			very small		1
			very small to small		2
			small	White Bush Scallop	3
			small to medium		4
			medium	Scallopini	5
			medium to large		6
			large	Sunburst	7
			large to very large		8
			very large		9

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note
33.2	33.2		<b><u>Only Acorn type varieties: Fruit:</u></b> <b>ratio length/ maximum diameter</b>		
			very small		1
			very small to small		2
			small	Heart of Gold	3
			small to medium		4
			medium	Table Gold	5
			medium to large		6
			large	Table King	7
			large to very large		8
			very large		9
33.3	33.3		<b><u>Only Zucchini type varieties: Fruit:</u></b> <b>ratio length/maximum diameter</b>		
			very small	Jericho	1
			very small to small		2
			small	Jedida	3
			small to medium		4
			medium	Cora	5
			medium to large		6
			large	Carlotta	7
			large to very large		8
			very large	Tarquinio	9



CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note
34.1	34.1 (*)		<b><u>Only Pumpkin type varieties: Fruit: size</u></b>		
			very small	Wee-B-Little	1
			very small to small		2
			small	Peek-a-Boo	3
			small to medium		4
			medium	Spirit	5
			medium to large		6
			large	Ghost Rider	7
			large to very large		8
			very large	Howden	9
34.2	34.2 (*)		<b><u>Only Rondini type varieties: Fruit: size</u></b>		
			very small		1
			very small to small		2
			small	Pomme d'Or	3
			small to medium		4
			medium	Rolet	5
			medium to large		6
			large	Little Gem	7
			large to very large		8
			very large		9
35.	35.		<b><u>Only Miniature Pumpkin type varieties: Fruit: peduncle end</u></b>		
			straight	Sweetie Pie	1
			concave	Jack Be Little	2

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note
36.	36.		<b><u>Only Scallop type varieties:</u> Fruit: protrusion of equatorial margin</b>		
			very weak		1
			very weak to weak		2
			weak	Scallopini	3
			weak to medium		4
			medium	Sunburst	5
			medium to strong		6
			strong	White Bush Scallop	7
			strong to very strong		8
			very strong		9
37.	37.		<b><u>Only Scallop type varieties:</u> Fruit: position of equatorial margin</b>		
			at the middle	Sunburst	1
			towards stem end	Golden Bush Scallop	2
38.	38.		<b><u>Only Scallop type varieties:</u> Fruit: peduncle end</b>		
			straight	White Bush Scallop	1
			convex	Yellow Bush Scallop	2
39.	39.		<b><u>Only Scallop type varieties:</u> Fruit: blossom end</b>		
			flat	Scallopini	1
			indented	Yellow Bush Scallop	2
40.	40.		<b><u>Only Acorn type varieties:</u> Fruit: position of maximum diameter</b>		
			at the middle	Ebony Table Queen	1
			towards stem end	Swan White Acorn	2
41.	41.		<b><u>Only Acorn type varieties:</u> Fruit: peduncle end</b>		
			concave	Swan White Acorn	1
			straight	Table King	2
			convex	Ebony Table Queen	3

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note
42.	42.		<b><u>Only Neck type varieties:</u> Fruit: length of neck</b>		
			short	Wryneck	3
			medium	Early Summer Crookneck	5
			long	Sundance	7
43.	43.		<b><u>Only Neck type varieties:</u> Fruit: diameter of neck in relation to maximum diameter</b>		
			very small		1
			very small to small		2
			small	Sundance	3
			small to medium		4
			medium	Early Summer Crookneck	5
			medium to large		6
			large	Sunbar	7
			large to very large		8
			very large		9
44.	44. (*)		<b><u>Only Neck type varieties:</u> Fruit: curving of neck</b>		
			absent	Early Prolific Straightneck	1
			present	Early Summer Crookneck	9
45.	45.		<b><u>Only Neck and Zucchini type varieties:</u> Fruit: blossom end</b>		
			rounded		1
			pointed		2
46.	46. (*)		<b>Fruit: grooves</b>		
			absent		1
			present		9

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note
47.	47.		<b>Fruit: depth of grooves</b>		
			very shallow	Spooktacular	1
			very shallow to shallow		2
			shallow	Connecticut Field	3
			shallow to medium		4
			medium	Delicata, Table Queen	5
			medium to deep		6
			deep	Jack Be Little, Swan White Acorn	7
			deep to very deep		8
			very deep	Heart of Gold, Sweet Dumpling	9
48.	48. (*)		<b>Fruit: ribs</b>		
			absent		1
			present		9
49.	49.		<b>Fruit: protrusion of ribs</b>		
			very weak	Leda, Tivoli	1
			very weak to weak		2
			weak	Ambassador	3
			weak to medium		4
			medium	Ibis, Opal	5
			medium to strong		6
			strong	Spidy	7
			strong to very strong		8
			very strong	Mogango Enrugado Mineiro, Tonda Padana	9

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note
50.	50. (*)		<b>Fruit: main colour of skin (excluding colour of dots, patches, stripes and bands)</b>		
			white	Pâtisson blanc panaché de vert	1
			cream	Early White Bush Scallop, Little Boo	2
			yellow	Autumn Gold	3
			green	Ambassador, Baby Bear	4
			partly white and partly yellow		5
			partly white and partly green		6
<b>G</b>			partly yellow and partly green	Sunburst, Zephyr	7
51.	51.		<b><u>Only varieties with yellow colour of skin: Fruit: intensity of yellow colour of skin (as for 50)</u></b>		
			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
52.	52.		<b><u>Only varieties with green colour of skin: Fruit: intensity of green colour of skin (as for 50)</u></b>		
			very light		1
			very light to light		2
			light		3
			light to meidum		4
			medium		5
			medium to dark		6
			dark	Cora	7
			dark to very dark		8
			very dark	Baby Bear, Sardane	9
53.	53.		<b><u>Varieties with two main colours only: Fruit: distribution of green colour</u></b>		
			green ring around blossom end	Sunburst	1
			from blossom end one third green	Zephyr	2
			from blossom end one half green		3
54. (+)	54.		<b>Fruit: stripes in grooves</b>		
			absent	Baby Bear, Jack Be Little	1
			present	Delicata, Heart of Gold, Pâtisson jaune panaché de vert	9
55.	55.		<b>Fruit: colour of stripes in grooves</b>		
			dark green	Sweet Dumpling	1
			yellow	Puccini	2
56. (+)	56.		<b>Fruit: colour of ribs compared to main colour of skin (as for 50)</b>		
			same	Grey Zucchini	1
			darker	De Nice à fruit rond, Orangetti	2

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note
57. (+)	57.		<b>Fruit: dots</b>		
			absent	Sunburst	1
			present	Gold Rush, Table Queen	9
58.	58.		<b>Fruit: size of main dots</b>		
			very small	Badger Cross	1
			very small to small		2
			small	Ambassador	3
			small to medium		4
			medium	Grey Zucchini	5
			medium to large		6
			large	Kingsize	7
			large to very large		8
			very large	Becky	9
59. (+)	59.		<b>Fruit: secondary green colour between ribs (excluding dots)</b>		
			absent	Grey Zucchini, Small Sugar	1
			present	Beatrice, Greyzini, Heart of Gold, Steierischer Ikürbis, Tonda Padana, Zubi	9
60. (+)	60.		<b>Fruit: intensity of secondary green colour between ribs</b>		
			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	Heart of Gold	9

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note
61. (+)	61.		<b>Fruit: distribution of secondary green colour between ribs</b>		
			sparse patches	Greyzini, Elite	1
			dense patches	Steierischer Ölkürbis	2
			one coloured stripes	Altea	3
			two coloured stripes		4
			one coloured bands covering the whole surface	Badger Cross, Twickers, Zubi	5
			two coloured bands covering the whole surface	Beatrice	6
62.	62. (*)		<b>Fruit: warts on skin</b>		
			absent		1
			present		9
63.	63.		<b>Fruit: number of warts on skin</b>		
			very few	Scallopini	1
			very few to few		2
			few		3
			few to medium		4
			medium	Pâtisson verruqueux panaché	5
			medium to many		6
			many	Early Summer Crookneck	7
			many to very many		8
			very many	Wryneck	9



CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note
<b>64.</b>	<b>64.</b>		<b>Fruit: size of flower scar</b>		
			very small	Jack be Little	1
			very small to small		2
			small	Goldi	3
			small to medium		4
			medium	Spidy	5
			medium to large		6
			large	Cinderella	7
			large to very large		8
			very large	Howden	9
<b>65.</b>	<b>65.</b>		<b>Fruit: length of peduncle</b>		
			very short	Arlesa	1
			very short to short		2
			short	Clarita	3
			short to medium		4
			medium	Cinderella	5
			medium to long		6
			long	Tivoli	7
			long ot very long		8
			very long	Western Sunrise	9
<b>66.</b>	<b>66.</b>		<b>Fruit: colour of peduncle</b>		
			yellow		1
			green	Ambassador	2
			partly yellow and partly green	Autumn Gold	3

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note
<b>67.</b>	<b>67.</b>		<b>Fruit: intensity of green colour of peduncle</b>		
			very light		1
			very light to light		2
			light	Bianchini	3
			light to medium		4
			medium	Sunburst	5
			medium to dark		6
			dark	Gold Rush	7
			dark to very dark		8
			very dark		9
<b>68.</b>	<b>68.</b>		<b>Fruit: mottling of peduncle</b>		
			absent	Sunburst	1
			present	Elite	9
<b>69. (+)</b>	<b>69. (*)</b>		<b>Ripe fruit: main colour of skin (excluding colour of mottles, patches, stripes and bands)</b>		
			white	Pâtisson blanc panaché de vert	1
			whitish	White Bush Scallop	2
			cream	Bianchini, Opal	3
			yellow	Gold Rush	4
			orange	Autumn Gold	5

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note
70.	70.		<b>Ripe fruit: intensity of main colour of skin (only yellow and orange)</b>		
			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
71.	71. (*)		<b>Ripe fruit: secondary colour of skin (as for 69)</b>		
			whitish		1
			cream		2
			yellow		3
			orange		4
			green		5
72.	72.		<b>Ripe fruit: green hue (only white and cream)</b>		
			absent	Jedida	1
			present	Amalthee	9

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note
<b>73.</b>	<b>73.</b>		<b>Ripe fruit: prominence of green hue (as for 72)</b>		
			very weak		1
			very weak to weak		2
			weak	Eskenderany	3
			weak to medium		4
			medium	Corona	5
			medium to strong		6
			strong	Amalthee	7
			strong to very strong		8
			very strong		9
<b>74.</b>	<b>74. (*)</b>		<b>Ripe fruit: colour of flesh</b>		
			cream	Elite	1
			yellow	Sunburst, Vegetable Spaghetti	2
			orange	Autumn Gold	3
<b>75.</b>	<b>75.</b>		<b>Ripe fruit: lignified rind</b>		
			absent	Small Sugar, Table Queen, Vegetable Spaghetti	1
			present	Elite, Little Gem, Scallopini, Early Summer Crookneck	9
<b>76.</b>	<b>76. (*)</b>		<b>Ripe fruit: structure of flesh</b>		
			not fibrous	Elite	1
			fibrous	Vegetable Spaghetti	2

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note
<b>77.</b>	<b>77.</b>		<b>Seed: size</b>		
			very small	Jack be Little	1
			very small to small		2
			small	Delicata	3
			small to medium		4
			medium	Diamant	5
			medium to large		6
			large		7
			large to very large		8
			very large	Citrouille de Touraine	9
<b>78.</b>	<b>78.</b>		<b>Seed: shape</b>		
			narrow elliptic	Caserta	3
			elliptic	Elite	5
			broad elliptic	Baby Boo	7
<b>79.</b>	<b>79.</b>		<b>Seed: hull</b>		
			absent	Chapingo Uno, Steierischer Ölkürbis	1
			present	Baby Bear, Elite	9
<b>80.</b>	<b>80.</b>		<b>Seed: appearance of hull</b>		
			rudimentary	Baby Bear	1
			fully developed	Elite	2
<b>81.</b>	<b>81.</b>		<b>Seed: colour of hull</b>		
			whitish	Table Queen	1
			cream	De Nice à fruit rond	2

CPVO N°	UPOV N°	Stage, Method	Characteristics	Examples	Note
<b>82. (+)</b>	<b>82.</b>	<b>VG/VS<sup>1</sup></b>	<b>Resistance to <i>Zucchini yellow mosaic virus</i> (ZYMV)</b>		
<b>QN<sup>2</sup></b>			absent or low	Cora	1
			medium	Mirza	2
			high	Mikonos	3
<b>83. (+)</b>	<b>83.</b>	<b>VG/VS<sup>1</sup></b>	<b>Resistance to <i>Watermelon mosaic virus</i> (WMV)</b>		
<b>QL<sup>2</sup></b>			absent	Cora	1
			present	Mikonos, Sofia, Syros	9

<sup>1</sup> See document TGP/7 "Development of Test Guidelines", Annex 3 "Guidance Notes (GN) for the TG Template", GN 25 "Recommendations for conducting the examination" ([http://www.upov.int/edocs/tgpdocs/en/tgp\\_7.pdf](http://www.upov.int/edocs/tgpdocs/en/tgp_7.pdf))

<sup>2</sup> See document TGP/7 "Development of Test Guidelines", Annex 3 "Guidance Notes (GN) for the TG Template", GN 20 "Presentation of characteristics: States of expression according to type of expression of a characteristic" ([http://www.upov.int/edocs/tgpdocs/en/tgp\\_7.pdf](http://www.upov.int/edocs/tgpdocs/en/tgp_7.pdf))

## 8. EXPLANATIONS ON THE TABLE OF CHARACTERISTICS

### 8.1 Explanations for individual characteristics

Ad. 13: Leaf blade: incisions



1  
absent or very shallow



3  
shallow



5  
Medium



7  
deep



9  
very deep

Ad. 26: Only Zucchini and Rounded Zucchini type varieties: Young fruit: general shape



1  
globular



2  
pear shaped



3  
tapered elliptical



4  
elliptical

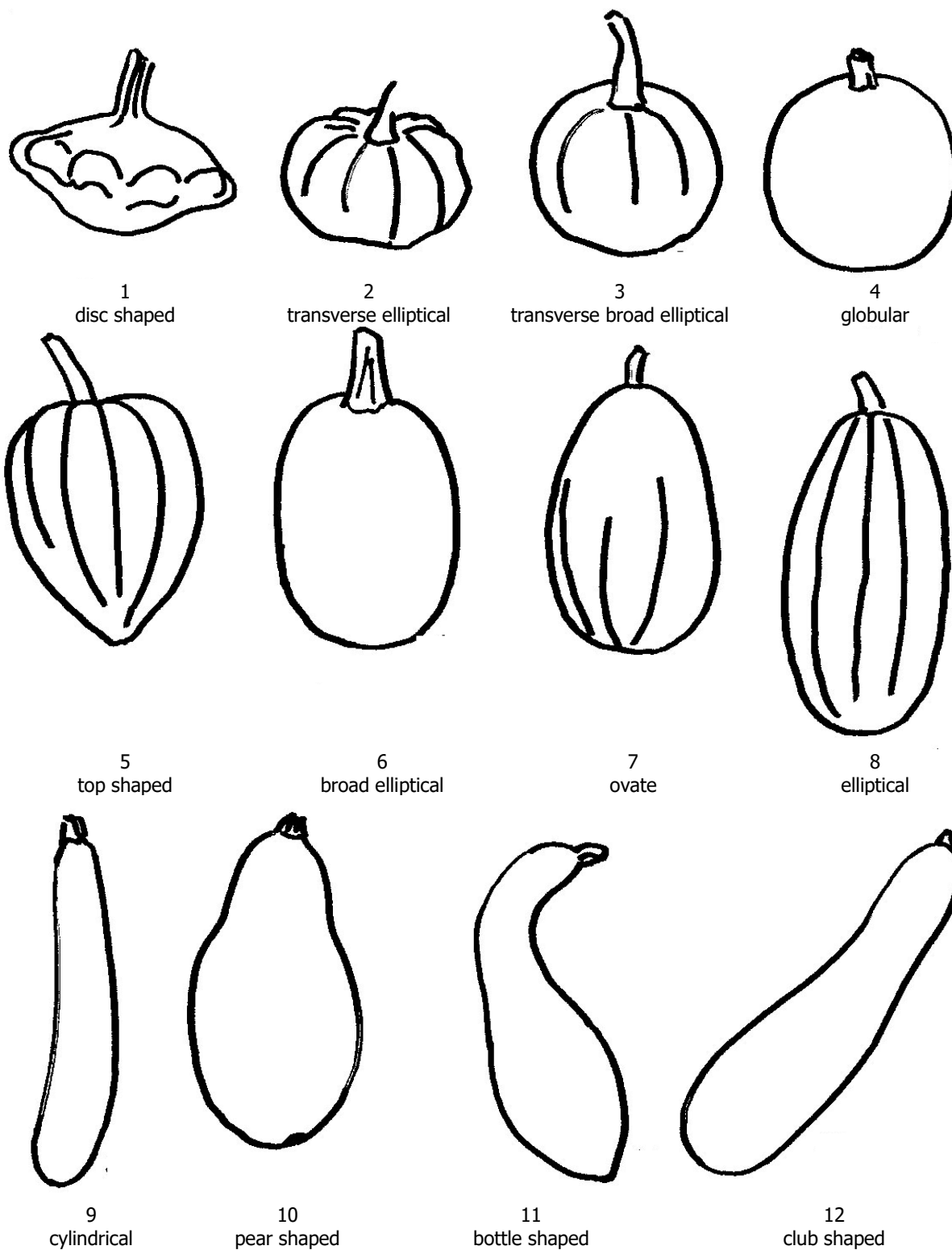


5  
cylindrical



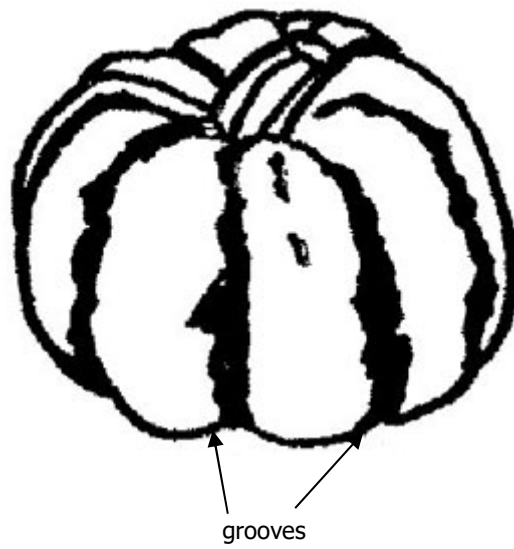
6  
tapered  
cylindrical

Ad. 30: Fruit: general shape

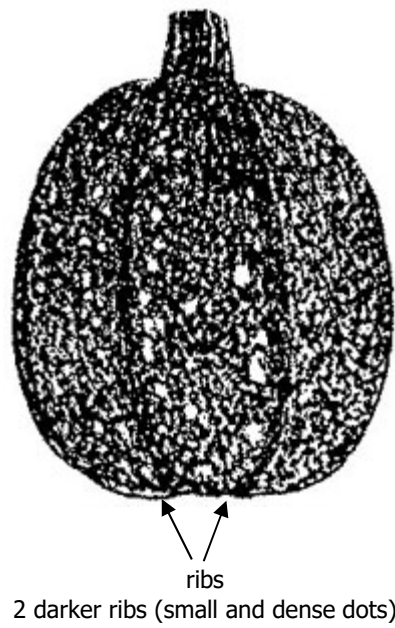




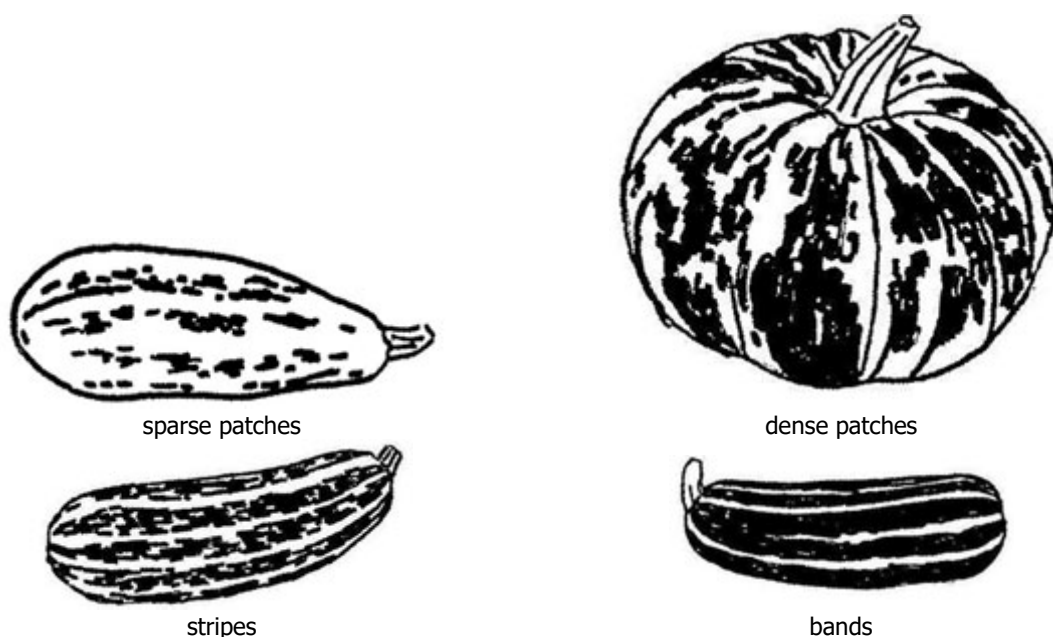
Ad. 54: Fruit: stripes in grooves



Ad. 56 and 57: Fruit: colour of ribs compared to main colour of skin (56), dots (57)



Ad. 59, 60, 61 and 69: Fruit: secondary colour between ribs (excluding dots)



Ad. 82: Resistance to *Zucchini yellow mosaic virus* (ZYMV)

1.	Pathogen	<i>Zucchini yellow mosaic virus</i> (ZYMV)
2.	Quarantine status	No
3.	Host species	<i>Cucurbita pepo</i> L.
4.	Source of inoculum	GEVES (FR) <sup>3</sup>
5.	Isolate	e.g. strain E9 = MAT/REF/06-08-02-02
6.	Establishment isolate identity	-
7.	Establishment pathogenicity	Symptoms on susceptible squash variety
8.	Multiplication inoculum	
8.1	Multiplication medium	Living plant
8.2	Multiplication variety	e.g. Cora
8.3	Plant stage at inoculation	-
8.4	Inoculation medium	-
8.5	Inoculation method	-
8.6	Harvest of inoculum	-
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 20
9.2	Number of replicates	At least 2 (e.g. 2x10 plants)
9.3	Control varieties	To illustrate and define the UPOV states <ul style="list-style-type: none"> <li>absent or low resistance (=susceptible): Cora</li> <li>medium resistance: Mirza (low threshold of medium resistance level):</li> <li>high resistance: Mikonos (low threshold of high resistance level)</li> </ul>
9.4	Test design	add non inoculated plants
9.5	Test facility	Climatic room or greenhouse
9.6	Temperature	e.g. 22°C or 24°C/18°C

<sup>3</sup> [matref@geves.fr](mailto:matref@geves.fr)

9.7	Light	12h-16h
9.8	Season	
9.9	Special measures	keep glasshouse free of aphids
10.	Inoculation	
10.1	Preparation inoculum	1 g leaf with symptoms with 4 mL of PBS with carborundum (400 mg) and activated carbon (400 mg) or similar buffer, homogenize
10.2	Quantification inoculum	
10.3	Plant stage at inoculation	First expanded leaf
10.4	Inoculation method	Rubbing cotyledons with virus suspension Optional: repeat the inoculation after 3 days
10.5	First observation	14 days post-inoculation
10.6	Second observation	-
10.7	Final observations	21 days post-inoculation
11.	Observations	
11.1	Method	Visual observation
11.2	Observation scale	Class 0: no symptom Class 1: few small chlorotic patches Class 2: many chlorotic patches Class 3: large chlorotic areas (some patches on young leaves) Class 4: mosaic and weak vein banding Class 5: deformation and vein banding



0: no symptom



1: few small chlorotic patches.



2: many chlorotic patches



3: large chlorotic areas (some patches on young leaves)



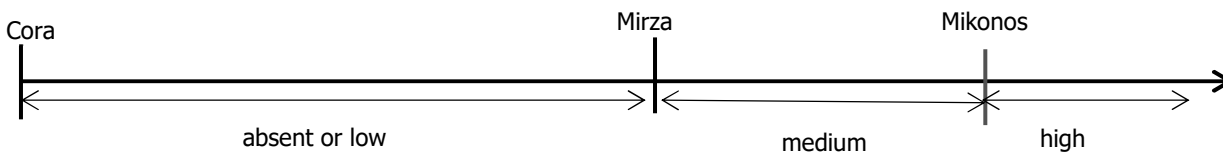
4: mosaic and weak vein banding



5: deformation and vein banding

Courtesy of GEVES-SNES







11.3	Validation of test	The high resistance control (Mikonos), the medium resistance control (Mirza) and the absent to low resistance (=susceptible) control (Cora) are necessary to validate the aggressiveness of test. Results should be compared with results of controls, based on disease index (DI) and distribution of plants over the classes.
11.4	Off-types	-

12.	Interpretation of data in terms of UPOV characteristic states	<p>- Note 1: Most plants are in class 4 and/or 5 (absent or low resistance = susceptible)</p> <p>- Note 2: Most plants are in class 2 and/or 3 (medium resistance)</p> <p>- Note 3: Most plants are in class 0 and/or 1 (high resistance)</p> <p>A variety with a lower resistance than Mirza (note 2) will be described note 1. A variety with a lower resistance than Mikonos (note 3), will be described note 2.</p> <p>An additional statistical analysis can be used to finalize the pathologist's raw observation to the assessment of uniformity, and relative position regarding the controls results.</p>
<p>Resistance to ZYMV:</p> 		
13.	Critical control points	<p>Date of notation may be adapted depending on expression of symptoms on controls.</p> <p>Environmental conditions can influence the expression of symptoms over time. In this case a second notation could be necessary.</p> <p>Aphids may transmit ZYMV as well as other viruses that may contaminate the ZYMV strain. Test should be in aphid-free compartment.</p>

#### Ad. 83: Resistance to *Watermelon mosaic virus* (WMV)

1.	Pathogen	<i>Watermelon mosaic virus</i> (WMV)
2.	Quarantine status	No
3.	Host species	<i>Cucurbita pepo</i> L.
4.	Source of inoculum	GEVES (FR) <sup>4</sup>
5.	Isolate	e.g., strain LL1A = MAT/REF/06-09-01 <sup>2</sup>
6.	Establishment isolate identity	-
7.	Establishment pathogenicity	Symptoms on susceptible squash variety
8.	Multiplication inoculum	
8.1	Multiplication medium	Living plant
8.2	Multiplication variety	e.g. Cora
8.3	Plant stage at inoculation	-
8.4	Inoculation medium	-
8.5	Inoculation method	-
8.6	Harvest of inoculum	-
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 20
9.2	Number of replicates	At least 2 (e.g. 2 x 10 plants)
9.3	Control varieties	<p>To illustrate UPOV notes:</p> <ul style="list-style-type: none"> <li>• resistance absent: Cora</li> <li>• resistance present: Sofia (minimum resistance level)</li> </ul> <p>Mikonos, Syros have higher resistance than Sofia, but not resistant enough to illustrate a high resistance.</p>
9.4	Test design	add non inoculated plants

<sup>4</sup> [matref@geves.fr](mailto:matref@geves.fr)

9.5	Test facility	Climatic room or greenhouse
9.6	Temperature	e.g., 22°C or 24°C/18°C
9.7	Light	12h-16h
9.8	Season	
9.9	Special measures	Keep glasshouse free of aphids
10.	Inoculation	
10.1	Preparation inoculum	1 g leaf with symptoms with 4mL of PBS with carborundum (400mg) and activated carbon (400mg) or similar buffer, homogenize
10.2	Quantification inoculum	-
10.3	Plant stage at inoculation	First expanded leave
10.4	Inoculation method	Rubbing cotyledons with virus suspension Optional: repeat the inoculation after 3 days
10.5	First observation	14 days post-inoculation
10.6	Second observation	-
10.7	Final observations	21 days post-inoculation
11.	Observations	
11.1	Method	Visual observation
11.2	Observation scale	Class 0: no symptom Class 1: few small chlorotic patches Class 2: many chlorotic patches Class 3: large chlorotic areas (some patches on young leaves) Class 4: mosaic, weak vein banding Class 5: deformation and vein banding
<div style="display: flex; flex-wrap: wrap; justify-content: space-around; align-items: flex-end;"> <div style="text-align: center;">  <p>0: no symptom</p> </div> <div style="text-align: center;">  <p>1: few small chlorotic patches</p> </div> <div style="text-align: center;">  <p>2: many chlorotic patches</p> </div> <div style="text-align: center;">  <p>3: large chlorotic areas (some patches on young leaves)</p> </div> <div style="text-align: center;">  <p>4: mosaic, weak vein banding</p> </div> <div style="text-align: center;">  <p>5: deformation and vein banding</p> </div> </div> <p style="text-align: right; margin-right: 50px;">Courtesy of GEVES-SNES</p>		
11.3	Validation of test	On three controls: Cora, Sofia, Mikonos or Syros The presence of Syros or Mikonos (and not only Sofia) is necessary to validate the aggressiveness of the test. Results should be compared with the results of controls, based on disease index (DI) and distribution of plants over the classes.
11.4	Off-types	-

12.	Interpretation of data in terms of UPOV characteristic states	<p>Note 1: Most plants are in class 4 and/or 5 (resistance absent or low to be considered)</p> <p>Note 9: Most plants are in class 0, 1, 2 and/or 3 (resistance present - more or less intensely)</p> <p>A variety with a lower level of resistance than Sofia (note 9), will be described as note 1</p> <p>An additional statistical analysis can be used to finalize the pathologist's raw observation to the assessment of uniformity, and relative position regarding the controls results.</p>
13.	Critical control points	<p>Date of notation may be adapted depending on expression of symptoms on controls.</p> <p>Environmental conditions can influence the expression of symptoms over time. In this case a second notation could be necessary.</p> <p>Aphids may transmit WMV as well as other viruses that may contaminate the WMV strain. Test should be in aphid-free compartment.</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-119/1-Rev.2 – *Cucurbita pepo* L. – vegetable marrow, squash