



EUROPEAN UNION

COMMUNITY PLANT VARIETY OFFICE

PROTOCOL FOR DISTINCTNESS, UNIFORMITY AND STABILITY TESTS

Citrus, Poncirus L. – Group 5

TRIFOLIATE ORANGE

UPOV Species Code: CITRU, PONCI_POL, PONCI_TRI

Adopted on 18/11/2004

I SUBJECT OF THE PROTOCOL

The protocol describes the technical procedures to be followed in order to meet the Council Regulation 2100/94 on Community Plant Variety Rights. The technical procedures have been agreed by the Administrative Council and are based on general UPOV Document TG/1/3 and UPOV guideline TG/83/4 dated 09/04/2003 for the conduct of tests for Distinctness, Uniformity and Stability. This protocol applies for all varieties of the following groups of the genus *Poncirus* L. (*Rutaceae*), and their hybrids. See below for the list of species concerned:

<i>Botanical taxon</i>	<i>Sub-group</i>	<i>Common name</i>
<i>Citrus clementina</i> Hort. ex Tan. x <i>Poncirus trifoliata</i> (L.) Raf.	HPO	Citrentin
<i>Citrus limon</i> (L.) Burm. x <i>Poncirus trifoliata</i> (L.) Raf.	CTL	Citremon
<i>Citrus paradisi</i> Macf. x <i>Poncirus trifoliata</i> (L.) Raf.	CML	Citrumelo
<i>Citrus reticulata</i> Blanco x <i>Poncirus trifoliata</i> (L.) Raf.	CTI	Citrandarin
<i>Citrus sinensis</i> (L.) Osb. x <i>Poncirus trifoliata</i> (L.) Raf.	CTG	Citrance
<i>Fortunella</i> sp. x <i>Poncirus trifoliata</i> (L.) Raf.	HPO	Citrumquat
<i>Poncirus polyandra</i> S. Q. Ding et al.	PON	
<i>Poncirus trifoliata</i> (L.) Raf.	PON	Golden Apple, Trifoliolate Orange

II SUBMISSION OF SEED AND OTHER PLANT MATERIAL

1. The Community Plant Variety Office (CPVO) is responsible for informing the applicant of
 - the closing date for the receipt of plant material;
 - the minimum amount and quality of plant material required;
 - the examination office to which material is to be sent.

A sub-sample of the material submitted for test will be held in the variety collection as the definitive sample of the candidate variety.

The applicant is responsible for ensuring compliance with any customs and plant health requirements.

2. Final dates for receipt of documentation and material by the Examination Office

The final dates for receipt of requests, technical questionnaires and the final date or submission period for plant material will be decided by the CPVO and each Examination Office chosen.

The Examination Office is responsible for immediately acknowledging the receipt of requests for testing, and technical questionnaires. Immediately after the closing date for the receipt of plant material the Examination Office should inform the CPVO whether acceptable plant material has been received or not. However if unsatisfactory plant material is submitted the CPVO should be informed as soon as possible.

3. Plant material requirements

The final dates for request for technical examination and sending of Technical Questionnaire by the CPVO as well as submission date, quantity and quality of plant material by the applicant can be found in the S2 supplement of the CPVO Official Gazette and the CPVO website (www.cpvo.europa.eu).

Quality of plants: As regards the health status, should not be less than the standards laid down in Council Directive 77/93/EEC, 92/34/EEC and 2000/29/EC. The plant material must be free from:

Insects, mites and nematodes at all stages of their development

- *Aleurothrixus floccosus* (Mashell)
- *Meloidogyne* spp.
- *Parabemisia myricae* (Kuwana)
- *Tylenchulus semipenetrans*

Fungi

- *Phytophthora* spp.

Viruses and virus-like organisms, and in particular

- Citrus leaf rugose
- Disease that induce psorosis-like young leaves symptoms such as: psorosis, ring pot cristicortis, impietratura, concave gum.
- Infectious variegation
- Viroids such as exocortis, cachexiaxyloporosis

Chemical treatment: The plant material must not have undergone any treatment unless the CPVO and the examination office allow or request such treatment. If it has been treated, full details of the treatment must be given.

Labelling of individual plants in sample:

- Species
- File number of the application allocated by the CPVO
- Breeder's reference
- Examination office's reference (if known)
- Name of applicant
- The phrase "On request of the CPVO"

III CONDUCT OF TESTS

1. Variety collection

A variety collection will be maintained for the purpose of establishing distinctness of the candidate varieties in test. A variety collection may contain both living material and descriptive information. A variety will be included in a variety collection only if plant material is available to make a technical examination.

Pursuant to Article 7 of Council Regulation No. 2100/94, the basis for a collection should be the following:

- varieties listed or protected at the EU level or at least in one of the EEA Member States;
- varieties protected in other UPOV Member States;
- any other variety in common knowledge.

The composition of the variety collection in each Examination Office depends on the environmental conditions in which the Examination Office is located.

Variety collections will be held under conditions which ensure the long term maintenance of each accession. It is the responsibility of Examination Offices to replace reference material which has deteriorated or become depleted. Replacement material can only be introduced if appropriate tests confirm conformity with the existing reference material. If any difficulties arise for the replacement of reference material, Examination Offices must inform the CPVO. If authentic plant material of a variety cannot be supplied to an Examination Office the variety will be removed from the variety collection.

2. Material to be examined

Candidate varieties will be directly compared with other candidates for Community plant variety rights tested at the same Examination Office, and with appropriate varieties in the variety collection. When necessary an Examination Office may also include other candidates and varieties. Examination Offices should therefore make efforts to co-ordinate the work with other Offices involved in DUS testing of trifoliolate orange. There should be at least an exchange of technical questionnaires for each candidate variety, and during the test period, Examination Offices should notify each other and the CPVO of candidate varieties which are likely to present problems in establishing distinctness. In order to solve particular problems Examination Offices may exchange plant material.

3. Characteristics to be used

The characteristics to be used in DUS tests and preparation of descriptions shall be those referred to in the Annex 1. 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. In the latter case, the CPVO should be informed. In addition the existence of some other regulation e.g. plant health, may make the observation of the characteristic impossible.

The Administrative Council empowers the President, in accordance with Article 23 of Commission Regulation N° 1239/95, to insert additional characteristics and their expression in respect of a variety.

4. Grouping of varieties

The varieties and candidates to be compared will be divided into groups to facilitate the assessment of distinctness. Characteristics which are suitable for grouping purposes are those which are known from experience not to vary, or to vary only slightly, within a variety and which in their various states of expression are fairly evenly distributed throughout the collection. In the case of continuous grouping characteristics overlapping states of expression between adjacent groups is required to reduce the risks of incorrect allocation of candidates to groups. The characters used for grouping could be the following:

- a) Leaf : caducity (characteristic 7)
- b) Leaf : number of leaflets (characteristic 8)
- c) Fruit surface: presence of pubescence (characteristic 72)
- d) Fruit: number of seeds (controlled manual self-pollination) (characteristic 100)
- e) Seed: polyembryony (characteristic 102)
- f) Self-incompatibility (characteristic 111)

5. Trial designs and growing conditions

The minimum duration of tests (independent growing cycles) will normally include at least two satisfactory crops of fruit. Tests will be carried out under conditions ensuring normal growth. The size of the plots will be such that plants or parts of plants may be removed for measuring and counting without prejudice to the observations which must be made up to the end of the growing period.

The test design is as follows

Each test should include 5 plants.

Unless otherwise indicated all observations determined by measuring and counting should be made 5 plants or 2 parts taken from each of 5 plants.

Young leaf: Observations on the young leaf should be made on actively growing spring flush.

Leaf: Observations on the leaf should be made on fully developed leaves on the middle third of the youngest spring flush branch sections not showing signs of active growth.

Flower: Unless otherwise indicated, observations on the flower bud and the flower should be made on the terminal flower bud and flower, at the time of full flowering of the variety.

Observations on the open flower should be made on the first day of opening.

Fruit: Observations on the fruit should be made at the stage of optimum ripeness. The fruit should be tested weekly and harvested as soon as this stage has been reached.

All fruits for observations should be taken from the periphery of the tree and fruit misformed as a result of clustering should not be sampled.

Fruit surface and fruit rind: Observations on the fruit surface and on the fruit rind should be made at the middle, between the base and apex of the fruit.

The observation on the oiliness of the fruit rind should be made, by peeling the fruit, within three to seven days after harvesting.

Fruit flesh: Observations on the flesh of the fruit should be made on a cross section through the middle of the fruit.

Seed: Observations on the seed should be made on the fresh seed.

6. Special tests

In accordance with Article 83(3) of Council Regulation No. 2100/94 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 characters listed in the protocol.

7. Standards for decisions

a) **Distinctness**

A candidate variety will be considered to be distinct if it meets the requirements of Article 7 of Council Regulation No. 2100/94.

b) **Uniformity**

A candidate will be considered to be sufficiently uniform if the number of off-types does not exceed the number of plants as indicated in the table below. A population standard of 1% and an acceptance probability of 95% should be applied.

Table of maximum numbers of off-types allowed for uniformity standards.

Number of plants	off-types allowed
≤ 5	0

c) **Stability**

A candidate will be considered to be sufficiently stable when there is no evidence to indicate that it lacks uniformity.

IV REPORTING OF RESULTS

After each recording season the results will be summarised and reported to the CPVO in the form of a UPOV model interim report in which any problems will be indicated under the headings distinctness, uniformity and stability. Candidates may meet the DUS standards after two fruiting periods but in some cases three fruiting periods may be required. When tests are completed the results will be sent by the Examination Office to the CPVO in the form of a UPOV model final report.

If it is considered that the candidate complies with the DUS standards, the final report will be accompanied by a variety description in the format recommended by UPOV. If not the reasons for failure and a summary of the test results will be included with the final report.

The CPVO must receive interim reports and final reports by the date agreed between the CPVO and the examination office.

Interim reports and final examination reports shall be signed by the responsible member of the staff of the Examination Office and shall expressly acknowledge the exclusive rights of disposal of CPVO.

V LIAISON WITH THE APPLICANT

If problems arise during the course of the test the CPVO should be informed immediately so that the information can be passed on to the applicant. Subject to prior agreement, the applicant may be directly informed at the same time as the CPVO particularly if a visit to the trial is advisable.

The interim report as well as the final report shall be sent by the Examination Office to the CPVO.

ANNEXES TO FOLLOW

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Legend:	
(+) See explanations on the Table of characteristics	
QL Qualitative characteristic	
QN Quantitative characteristic	
PQ Pseudo-qualitative characteristic	
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ANNEX II

Technical Questionnaire

ANNEX I

TABLE OF CHARACTERISTICS TO BE USED IN DUS-TEST AND PREPARATION OF DESCRIPTIONS

CPVO N°	UPOV N°	Characteristics	Examples	Note	
1. (+) PQ	1.	Tree: growth habit	upright	<i>Poncirus trifoliata</i> (PON)	1
			spreading	CPB 4475 (CML)	2
			drooping		3
2. QN	2.	Tree: density of spines	absent or sparse		1
			intermediate		2
			dense	<i>Poncirus trifoliata</i> (PON)	3
3. QN	3.	Tree: length of spines	short		3
			medium		5
			long	<i>Poncirus trifoliata</i> (PON)	7
4. QN	4.	Branch: expression of lenticels	weak		3
			medium		5
			strong	Cunningham (CTG)	7
5. QL	5.	Young leaf: presence of anthocyanin coloration	absent	Troyer (CTG)	1
			present		9
6. QN	6.	Young leaf: intensity of anthocyanin coloration	weak		3
			medium		5
			strong		7
7. PQ	7.	Leaf: caducity	perennial	CPB 4475 (CML)	1
			partially caducous	Troyer (CTG)	2
			caducous	<i>Poncirus trifoliata</i> (PON)	3
8. PQ	8.	Leaf: number of leaflets	one only		1
			variable	Troyer (CTG)	2
			three only	<i>Poncirus trifoliata</i> (PON)	3

CPVO N°	UPOV N°	Characteristics	Examples	Note
9. QN	9.	Leaf blade: length (apical leaflet in case of compound leaf)	short	3
			medium	5
			long	7
10. QN	10.	Leaf blade: width (apical leaflet in case of compound leaf)	narrow	3
			medium	5
			broad	7
11. QN	11.	Leaf blade: ratio length/width (apical leaflet in case of compound leaf)	small	<i>Poncirus trifoliata</i> (PON) 3
			medium	5
			large	7
12. QN	12.	Leaf blade: length (lateral leaflet in case of compound leaf)	short	3
			medium	5
			long	7
13. QN	13.	Leaf blade: width (apical leaflet in case of compound leaf)	narrow	3
			medium	5
			broad	7
14. QN	14.	Leaf blade: ratio length/width (apical leaflet in case of compound leaf)	small	<i>Poncirus trifoliata</i> (PON) 3
			medium	5
			large	7
15. QN	15.	Leaf blade: ratio length of blade of apical leaflet/length of blade of lateral leaflet	small	3
			medium	5
			large	7

CPVO N°	UPOV N°	Characteristics	Examples	Note
16. QN	16.	Leaf blade: shape in cross section (apical leaflet in case of compound leaf)	straight or weakly concave	1
			intermediate	2
			strongly concave	3
17. QN	17.	Leaf blade: twisting	absent or weak	1
			intermediate	2
			strong	3
18. QN	18.	Leaf blade: blistering	absent or weak	1
			intermediate	2
			strong	3
19. QN	19.	Leaf blade: green colour	light	3
			medium	5
			dark	7
20. QN	20.	Leaf blade: pubescence on lower side	absent or weak	1
			intermediate	2
			strong	3
21. QN	21.	Leaf blade: undulation of margin	absent or weak	1
			intermediate	2
			strong	3
22. PQ	22.	Leaf blade: incisions of margin	absent	1
			crenate	2
			dentate	3
23. (+) PQ	23.	Leaf blade: shape of apex	acuminate	1
			acute	2
			obtuse	3
			rounded	4

CPVO N°	UPOV N°	Characteristics	Examples	Note
24. (+) QL	24.	Leaf blade: emargination at tip	absent	1
			present	9
25. QN	25.	Petiole: length	short	3
			medium	5
			long	7
26. QL	26.	Petiole: presence of wings	absent	1
			present	9
27. QN	27.	<u>Varieties with petiole wings present only:</u> Petiole: width of wings	narrow	3
			medium	5
			broad	7
28. QL	28.	Flower bud: presence of anthocyanin coloration	absent	1
			present	9
29. QN	29.	Flower bud: intensity of anthocyanin coloration	weak	3
			medium	5
			strong	7
30. QN	30.	Flower: diameter of calyx	small	3
			medium	5
			large	7
31. QN	31.	Flower: length of petal	short	3
			medium	5
			long	7
32. QN	32.	Flower: width of petal	narrow	3
			medium	5
			broad	7

CPVO N°	UPOV N°	Characteristics		Examples	Note
33. QN	33.	Flower: ratio length/width of petal	small		3
			medium		5
			large		7
34. QN	34.	Flower: length of stamens	short		3
			medium		5
			long		7
35. QL	35.	Stamen: colour of filament	white	Carrizo (CTG)	1
			pink	Forner Alcaide 13 (CTI)	2
36. PQ	36.	Anther: colour	white		1
			light yellow		2
			medium yellow		3
37. QL	37.	Anther: viable pollen	absent		1
			present		9
38. QN	38.	Style: length	short		3
			medium		5
			long		7
39. QL	39.	Ovary: pubescence	absent		1
			present	<i>Poncirus trifoliata</i> (PON)	9
40. QN	40.	Fruit: length	short		3
			medium		5
			long		7
41. QN	41.	Fruit: diameter	small		3
			medium		5
			large		7
42. QN	42.	Fruit: ratio length/diameter	small		3
			medium		5
			large		7

CPVO N°	UPOV N°	Characteristics	Examples	Note	
43. QN	43.	Fruit: position of broadest part	towards stalk end	1	
			at middle	2	
			towards distal end	3	
44. (+) PQ	44.	Fruit: shape in transverse section	circular	<i>Poncirus trifoliata</i> (PON)	1
			somewhat angular		2
			scalloped		3
45. (+) PQ	45.	Fruit: general shape of proximal part (excluding neck, collar and depression at stalk end)	flattened		1
			slightly rounded		2
			strongly rounded		3
			tapered		4
46. (+) QL	46.	Fruit: presence of neck	absent		1
			present		9
47. QN	47.	<u>Necked varieties only:</u> Fruit: length of neck	short		3
			medium		5
			long		7
48. QN	48.	<u>Necked varieties only:</u> Fruit: thickness of neck	thin		3
			medium		5
			thick		7
49. QL	49.	<u>Only varieties without fruit neck:</u> Fruit: presence of depression at stalk end	absent		1
			present		9
50. QN	50.	<u>Only varieties without fruit neck:</u> Fruit: depth of depression at stalk end	shallow		3
			medium		5
			deep		7

CPVO N°	UPOV N°	Characteristics	Examples	Note
51. QL	51.	Fruit: presence of constriction at stalk end	absent	1
			present	9
52. QN	52.	Fruit: expression of constriction at stalk end	weak	3
			medium	5
			strong	7
53. QN	53.	Fruit: number of radial grooves at stalk end	absent or few	1
			intermediate	2
			many	3
54. QN	54.	Fruit: length of radial grooves at stalk end	short	3
			medium	5
			long	7
55. QN	55.	<u>Necked varieties only</u> : Fruit: depression at stalk attachment	absent or shallow	1
			intermediate	2
			deep	3
56. (+) QL	56.	Fruit: presence of collar	absent	1
			present	9
57. QN	57.	Fruit: abscission layer between floral disc and fruit	absent or weakly developed	1
			intermediate	2
			strongly developed	3
58. (+) QN	58.	Fruit: general shape of distal part (excluding nipple, bulging of navel and depression at distal end)	flattened	1
			slightly rounded	2
			strongly rounded	3

CPVO N°	UPOV N°	Characteristics	Examples	Note
59. (+) QL	59.	Fruit: presence of depression at distal end	absent	1
			present	9
60. QN	60.	Fruit: depth of depression at distal end	shallow	3
			medium	5
			deep	7
61. (+) QL	61	Fruit: presence of nipple	absent	1
			present	9
62. QN	62.	Fruit: prominence of nipple	weak	3
			medium	5
			strong	7
63. QL	63.	Fruit: presence of areola	absent	1
			incomplete	2
			complete	3
64. QL	64.	Fruit: type of areola	smooth	1
			grooved	2
			ridged	3
65. QN	65.	Fruit: diameter of areola	small	3
			medium	5
			large	7
66. QN	66.	Fruit: diameter of stylar scar	small	3
			medium	5
			large	7
67. QN	67.	Fruit: protruding stylar point	small	3
			medium	5
			large	7

CPVO N°	UPOV N°	Characteristics	Examples	Note
68. PQ	68.	Fruit: persistence of style	none	1
			partial	2
			total	3
69. QN	69.	Fruit: bulging of navel	absent or weak	1
			intermediate	2
			strong	3
70. QL	70.	Fruit: presence of radial grooves at distal end	absent	1
			present	9
71. PQ	71.	Fruit surface: predominant colour(s)	green	1
			yellow green	2
			green and yellow	3
			light yellow	4
			medium yellow	5
			yellow orange	6
			medium orange	7
			dark orange	8
			green and orange	9
			yellow and orange	10
			orange red	11
			yellow and red	12
			orange and red	13
72. QL	72.	Fruit surface: presence of pubescence	absent	1
			present	<i>Poncirus trifoliata</i> (PON) 9
73. QN	73.	Fruit surface: intensity of pubescence	weak	3
			medium	5
			strong	7

CPVO N°	UPOV N°	Characteristics	Examples	Note
74. QN	74.	Fruit surface: roughness	smooth	3
			medium	5
			rough	7
75. PQ	75.	Fruit surface: size of oil glands	all more or less the same size	1
			larger ones interspersed by smaller ones	2
76. QN	76.	Fruit surface: size of larger oil glands	small	3
			medium	5
			large	7
77. QN	77.	Fruit surface: conspicuousness of larger oil glands	weak	3
			medium	5
			strong	7
78. PQ	78.	Fruit surface: presence of pitting and pebbling on oil glands	pitting and pebbling absent	1
			pitting absent, pebbling present	2
			pitting present, pebbling absent	3
			pitting and pebbling present	4
79. QN	79.	<u>Varieties with fruit surface: pitting on oil glands present only:</u> Fruit surface: density of pitting	sparse	3
			medium	5
			dense	7

CPVO N°	UPOV N°	Characteristics	Examples	Note
80.	80.	<u>Varieties with fruit surface: pitting on oil glands present only:</u> Fruit surface: depth of pitting	shallow	3
QN			medium	5
			deep	7
81.	81.	<u>Varieties with fruit surface: pebbling on oil glands present only:</u> Fruit surface: density of pebbling	sparse	3
QN			medium	5
			dense	7
82.	82.	<u>Varieties with fruit surface: pebbling on oil glands present only:</u> Fruit surface: degree of pebbling	weak	3
QN			medium	5
			strong	7
83.	83.	Fruit rind: thickness	thin	3
QL			medium	5
			thick	7
84.	84.	Fruit rind: adherence to flesh	weak	3
QN			medium	5
			strong	7
85.	85.	Fruit rind: strength	weak	3
QN			medium	5
			strong	7
86.	86.	Fruit rind: conspicuousness of oil glands on inner surface	absent or weakly conspicuous	1
QN			intermediate	2
			strongly conspicuous	3

CPVO N°	UPOV N°	Characteristics	Examples	Note
87. PQ	87.	Fruit: colour of albedo	greenish	1
			white	2
			light yellow	3
			light orange	4
			pink	5
			reddish	6
88. QN	88.	Fruit: density of albedo	loose	3
			medium	5
			dense	7
89. QN	89.	Fruit: amount of albedo adhering to flesh (strands excluded)	absent or very small	1
			small	3
			medium	5
			large	7
			very large	9
90. QL	90.	Fruit: presence of albedo strands	absent	1
			present	9
91. QN	91.	Fruit: amount of albedo strands	small	3
			medium	5
			large	7

CPVO N°	UPOV N°	Characteristics	Examples	Note
92. PQ	92.	Fruit: main colour of flesh	whitish	1
			light green	2
			light yellow	3
			medium yellow	4
			light orange	5
			medium orange	6
			dark orange	7
			red	8
			purple	9
			yellow and red	10
93. QL	93.	Fruit: bitterness of flesh	absent	1
			present	9
94. QN	94.	Fruit: filling of core	absent or very sparse	1
			sparse	3
			medium	5
			dense	7
95. QN	95.	Fruit: presence of rudimentary segments	absent or weak	1
			intermediate	2
			strong	3
96. QN	96.	Fruit: number of well developed segments	few	3
			medium	5
			many	7
97. QN	97.	Fruit: strength of segment walls	weak	3
			medium	5
			strong	7

CPVO N°	UPOV N°	Characteristics	Examples	Note
98. PQ	98.	Fruit: presence of navel (viewed internally)	absent or very rare	1
			occasionally present	2
			always present	3
99. QN	99.	Fruit: juiciness	low	3
			medium	5
			high	7
100. (+) QN	100.	Fruit: number of seeds (controlled manual self-pollination)	absent or very few	1
			few	3
			medium	5
			many	7
			very many	9
101. (+) QN	101	Fruit: number of seeds (open pollination)	absent or very few	1
			few	3
			medium	5
			many	7
102. QL	102.	Seed: polyembryony	absent	1
			present	9
103. QN	103.	Seed: length	short	3
			medium	5
			long	7
104. QN	104.	Seed: width	narrow	3
			medium	5
			broad	7
105. QL	105.	Seed: surface	smooth	1
			wrinkled	2

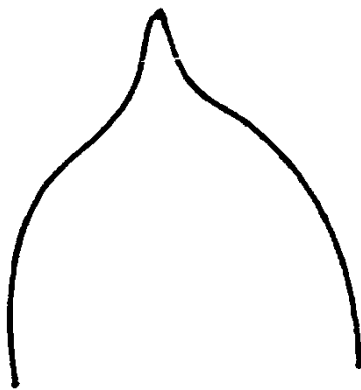
CPVO N°	UPOV N°	Characteristics	Examples	Note
106. PQ	106.	Seed: external colour	greenish	1
			whitish	2
			yellowish	3
			pinkish	4
			brownish	5
107. PQ	107.	Seed: colour of inner seed coat	white	1
			light yellow	2
			light brown	3
			medium brown	4
			dark brown	5
			red	6
			purple	7
108. PQ	108.	<u>Only varieties with seed: polyembryony present:</u> Seed: colour of cotyledons	white	1
			cream	2
			light	3
			dark green	4
109. QL	109.	Flowering habit	flowering once	1
			flowering more than once	2
110. QN	110.	Time of maturity of fruit for consumption	early	3
			medium	5
			late	7
111. (+) QL	111.	Self-incompatibility	absent	1
			present	9

EXPLANATIONS AND METHODS

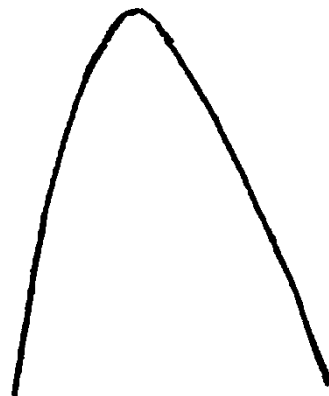
Ad. 1 : Tree: Growth habit

The observation on the growth habit of the tree should be made immediately after harvest.

Ad. 23 : Leaf blade: shape of apex



1
acuminate



2
acute

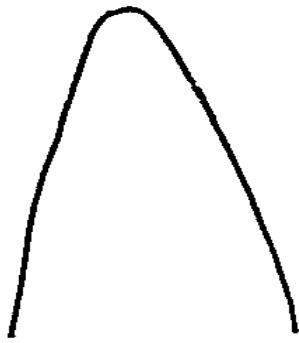


3
obtuse

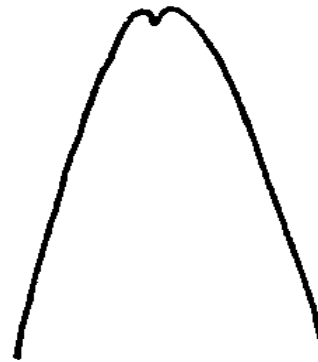


4
rounded

Ad. 24 : Leaf blade: emargination at tip

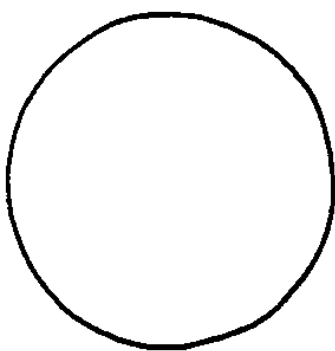


1
absent

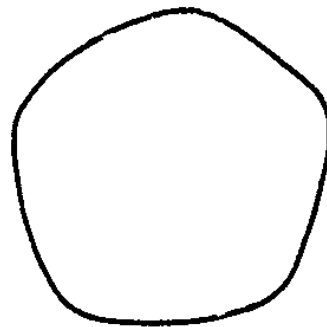


9
present

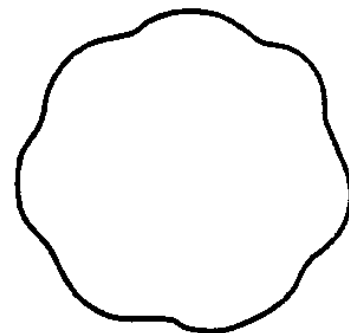
Ad. 44 : Fruit: shape in transverse section



1
circular

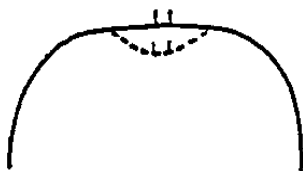


2
somewhat angular

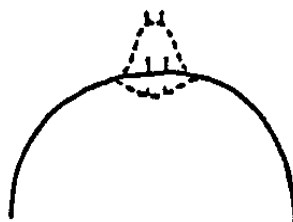


3
scalloped

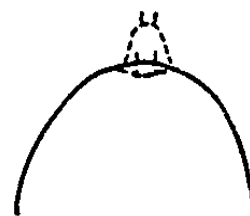
Ad. 45 : Fruit: general shape of proximal part (excluding neck, collar and depression at stalk end)



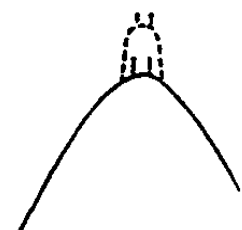
1
flattened



2
slightly rounded



3
strongly rounded

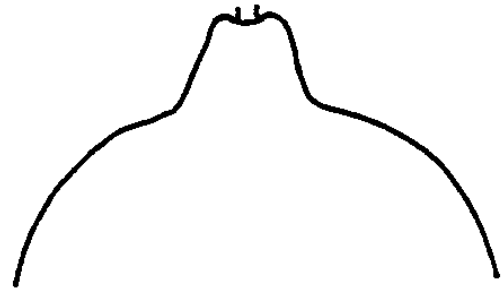


4
tapered

Ad. 46 : Fruit: presence of neck

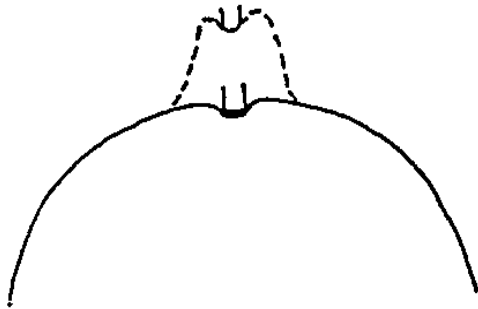


1
absent

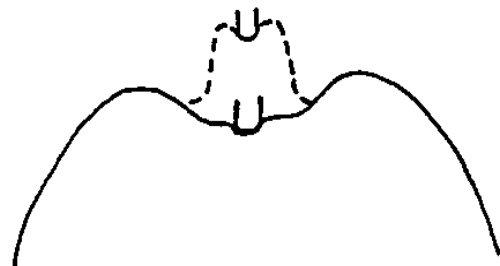


9
present

Ad. 49 : Only varieties without fruit neck: Fruit: presence of depression at stalk end

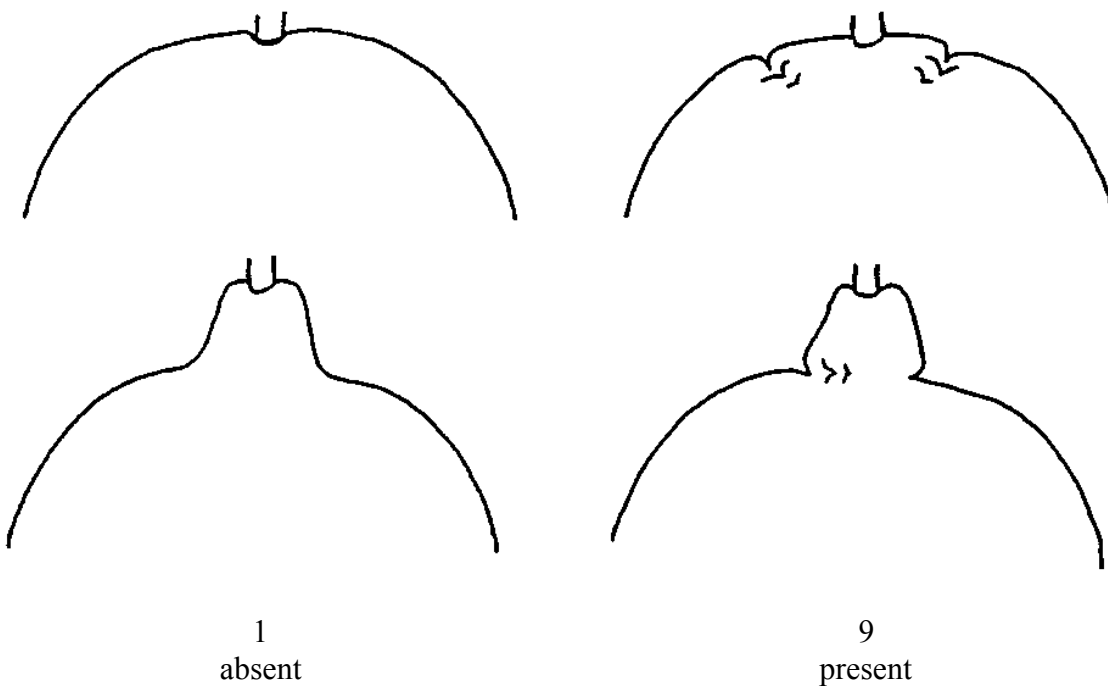


1
absent

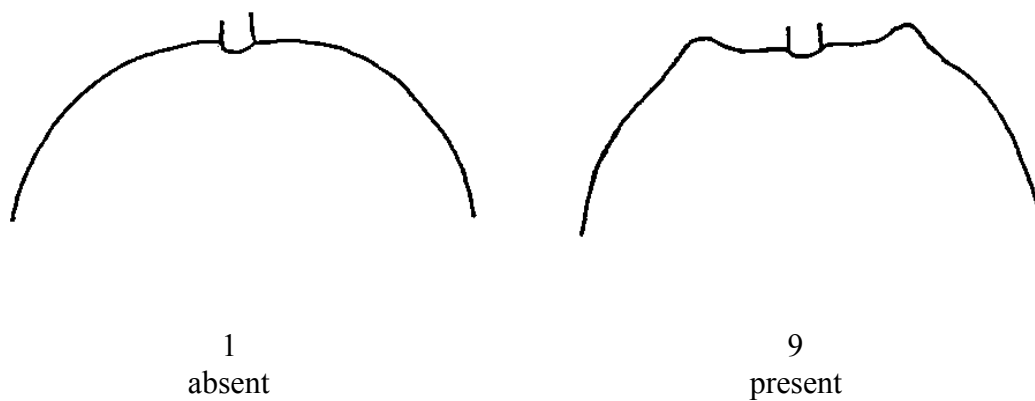


9
present

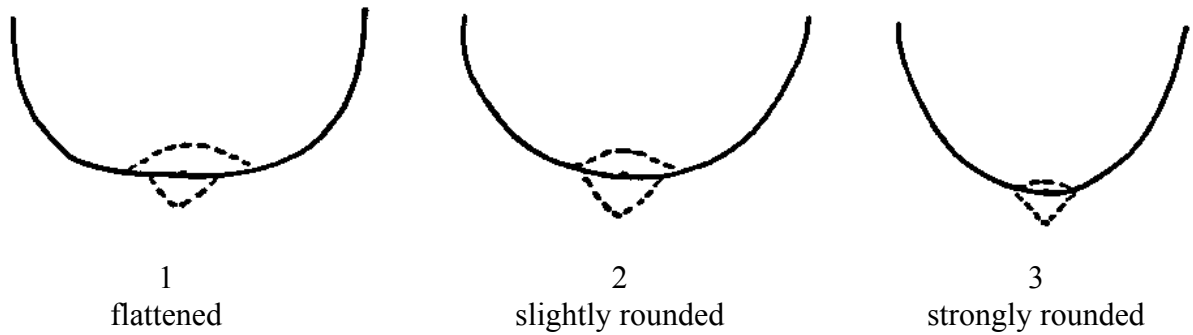
Ad. 51: Fruit: presence of constriction at stalk end



Ad. 56 : Fruit: presence of collar



Ad. 58 : Fruit: general shape of distal part (excluding nipple, bulging of navel and depression at distal end)



Ad 59 : Fruit: presence of depression at distal end



Ad. 61 : Fruit: presence of nipple



Ad. 64 : Fruit: type of areola



1
smooth



2
grooved



3
ridged

Ad. 100 : Fruit: number of seeds (controlled manual self-pollination)

Manual self-pollination is necessary to ensure a consistent production of seed.

Ad 101 : Fruit: number of seeds (open pollination)

Open pollination means natural pollination between trees of the same variety.

Ad. 111 : Self-incompatibility

A variety is self-incompatible when the fertile pollen of its own flower or of other flowers of the same variety is not able to fertilize the ovary.

The test on self-incompatibility has to be carried out on at least 10 flowers.

Choose flowers with petals which are just before opening and open the flower manually. Then separate and cut the anthers. Take viable pollen from other flowers of the same variety and put it on the stigma. Cover the flowers with muslin in order to avoid accidental pollination by other pollen. If the mature fruit bears no seeds, the variety is self-incompatible. If the mature fruit bears seeds, the variety is self-compatible.

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ANNEX II



European Union
Community Plant Variety Office

TECHNICAL QUESTIONNAIRE

to be completed in connection with an application for Community Plant Variety Rights
Please answer all questions. A question without any answer will lead to a non-attribution
of an application date. In cases where a field / question is not applicable, please state so.

1. **Botanical taxon:** Name of the genus, species or sub-species to which the variety belongs and common name

Citrus, Poncirus L. – Group 5

TRIFOLIATE ORANGE

2. **Applicant(s):** Name(s) and address(es), phone and fax number(s), Email address, and where appropriate name and address of the procedural representative

3. **Variety denomination**

a) Where appropriate proposal for a variety denomination:

b) Provisional designation (breeder's reference):

4. Information on origin, maintenance and reproduction of the variety

4.1 Breeding, maintenance and reproduction of the variety

Please indicate breeding scheme, parents and other relevant information.

- (a) Seedling of unknown parentage[]
- (b) Produced by controlled pollination (indicate parent varieties).....[]
 - (i) Seed bearing parent

 - (ii) Pollen parent
- (c) Produced by open pollination of (indicate seed bearing parent only)[]
- (d) Mutation or sport from (indicate parent variety)[]
- (d) Discovery (indicate where and when)[]

4.2 Method of propagation

- (a) Cuttings[]
- (b) *In vitro* propagation.....[]
- (c) Seed[]
- (d) Other (please specify):[]

4.3 Virus status

The variety is:

- (i) Virus free (indicate viruses).....[]
- (ii) Virus tested (indicate against which virus)[]
- (iii) The virus status is unknown.....[]

4.4 Geographical origin of the variety: the region and the country in which the variety was bred or discovered and developed

5. Characteristics of the variety to be indicated (the number in brackets refers to the corresponding characteristic in the CPVO Protocol; please mark the state of expression which best corresponds).

	Characteristics	Example varieties	Note
5.1 (7)	Leaf: caducity		
	perennial	CPB 4475 (CML)	1 []
	partially caducous	Troyer (CTG)	2 []
	caducous	<i>Poncirus trifoliata</i> (PON)	3 []
5.2 (8)	Leaf: number of leaflets		
	one only		1 []
	variable	Troyer (CTG)	2 []
	three only	<i>Poncirus trifoliata</i> (PON)	3 []
5.3 (72)	Fruit surface: presence of pubescence		
	absent		1 []
	present	<i>Poncirus trifoliata</i> (PON)	9 []

Characteristics		Example varieties	Note
5.4 (100)	Fruit : number of seeds (controlled manual self-pollination)		
	absent or very few		1 []
	few		3 []
	medium		5 []
	many		7 []
	very many		9 []
5.5 (102)	Seed: polyembryony		
	absent		1 []
	preent		9 []
5.6 (111)	Self-incompatibility		
	absent		1 []
	present		9 []
6. Similar varieties and differences from these varieties:			
Denomination of similar variety	Characteristic in which the similar variety is different ¹⁾	State of expression of similar variety	State of expression of candidate variety
<p>¹⁾ In the case of identical states of expressions of both varieties, please indicate the size of the difference</p>			
7. Additional information which may help to distinguish the variety			
A representative printed-out colour photo of the variety must be added to the Technical Questionnaire.			
7.1 Resistance to pests and diseases			

7.2 Special conditions for the examination of the variety

YES, please specify

NO

7.3 Other information

YES, please specify

NO

8. GMO-information required

The variety represents a Genetically Modified Organism within the meaning of Article 2(2) of Council Directive EC/2001/18 of 12/03/2001.

YES NO

If yes, please add a copy of the written attestation of the responsible authorities stating that a technical examination of the variety under Articles 55 and 56 of the Basic Regulation does not pose risks to the environment according to the norms of the above-mentioned Directive.

9. Information on plant material to be examined

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

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

- | | | |
|---|------------------------------|-----------------------------|
| (a) Microorganisms (e.g. virus, bacteria, phytoplasma) | <input type="checkbox"/> Yes | <input type="checkbox"/> No |
| (b) Chemical treatment (e.g. growth retardant or pesticide) | <input type="checkbox"/> Yes | <input type="checkbox"/> No |
| (c) Tissue culture | <input type="checkbox"/> Yes | <input type="checkbox"/> No |
| (d) Other factors | <input type="checkbox"/> Yes | <input type="checkbox"/> No |

Please provide details of where you have indicated "Yes":

I/we hereby declare that to the best of my/our knowledge the information given in this form is complete and correct.

Date

Signature

Name

[End of document]