

ANNEX III

QUARANTINE MEASURES INCLUDING TESTING ON PLANTS, PLANT PRODUCTS AND OTHER OBJECTS INTENDED FOR RELEASE FROM QUARANTINE

PART A

For certain plants, plant products and other objects listed in Annex III to Directive 2000/29/EC

Section I:

Plants of *Citrus* L., *Fortunella* Swingle, *Poncirus* Raf. and their hybrids, other than fruit and seeds

1. The plant material, as appropriate, shall be subjected to appropriate therapy procedures as laid down in FAO/IPGRI Technical Guidelines.
2. The plant material, following the therapy procedures carried out pursuant to point 1, shall be subjected to indexing procedures in its entirety. All plant material including indexing plants, shall be held at the approved facilities under the quarantine containment conditions laid down in Annex I. Plant material intended for approval for official release shall be held under conditions conducive to a normal cycle of vegetative growth and be subjected to visual inspection for signs and symptoms of harmful organisms including all relevant harmful organisms listed in Directive 2000/29/EC, on arrival and subsequently, at appropriate times, during the period of the indexing procedures.
3. For the purposes of point 2, the plant material shall be indexed for harmful organisms (tested for and identified) according to the following procedures:
 - 3.1. The testing shall use appropriate laboratory methods and, where appropriate, indicator plants, including *Citrus sinensis* (L.) Osbeck, *C. aurantiifolia* Christm. Swing, *C. medica* L., *C. reticulata* Blanco and *Sesamum* L., in order to detect at least the following harmful organisms:
 - (a) Citrus greening bacterium
 - (b) Citrus variegated chlorosis
 - (c) Citrus mosaic virus
 - (d) Citrus tristeza virus (all isolates)
 - (e) Citrus vein enation woody gall
 - (f) Leprosis
 - (g) Naturally spreading psorosis
 - (h) *Phoma tracheiphila* (Petri) Kanchaveli & Gikashvili
 - (i) Satsuma dwarf virus
 - (j) *Spiroplasma citri* Saglio *et al.*

- (k) Tatter leaf virus
 - (l) Witches' broom (MLO)
 - (m) *Xanthomonas campestris* (all strains pathogenic to *Citrus*).
- 3.2. For diseases such as Blight and Blight-like for which there are no short-term indexing procedures the plant material must be subjected upon arrival to shoot-tip grafting onto seedling stock grown under sterile culture as set out in FAO/IPGRI Technical Guidelines, and the resulting plants subjected to therapy procedures according to point 1.
4. The plant material subjected to the visual inspections referred to in point 2 and on which signs and symptoms of harmful organisms have been observed shall be subjected to an investigation including testing where necessary, to determine, as far as possible, the identity of the harmful organisms causing the signs and symptoms.

Section II:

Plants of *Cydonia* Mill., *Malus* Mill., *Prunus* L. and *Pyrus* L. and their hybrids and *Fragaria* L., intended for planting, other than seeds

1. The plant material, as appropriate, shall be subjected to appropriate therapy procedures as laid down in FAO/IPGRI Technical Guidelines.
2. The plant material, following the therapy procedures carried out pursuant to point 1, shall be subjected to indexing procedures in its entirety. All plant material including indexing plants, shall be held at the approved facilities under the quarantine containment conditions laid down in Annex I. Plant material intended for approval for official release shall be held under conditions conducive to a normal cycle of vegetative growth and be subjected to visual inspection for signs and symptoms of harmful organisms including all relevant harmful organisms listed in Directive 2000/29/EC, on arrival and subsequently, at appropriate times, during the period of the indexing procedures.
3. For the purposes of point 2 the plant material shall be indexed for harmful organisms (tested for and identified) according to the following procedures:
 - 3.1. In the case of *Fragaria* L., irrespective of the country of origin of the plant material, the testing shall use appropriate laboratory methods and, where appropriate, indicator plants, including *Fragaria vesca*, *F. virginiana* and *Chenopodium* spp. for the detection of at least the following harmful organisms:
 - (a) Arabis mosaic virus
 - (b) Raspberry ringspot virus
 - (c) Strawberry crinkle virus
 - (d) Strawberry latent 'C' virus
 - (e) Strawberry latent ringspot virus
 - (f) Strawberry mild yellow edge virus
 - (g) Strawberry vein banding virus

- (h) Strawberry witches' broom mycoplasma
- (i) Tomato black ring virus
- (j) Tomato ringspot virus
- (k) *Colletotrichum acutatum* Simmonds
- (l) *Phytophthora fragariae* Hickman var. *fragariae* Wilcox & Duncan
- (m) *Xanthomonas fragariae* Kennedy & King.

3.2. In the case of *Malus* Mill.:

- (i) where the plant material originates from a country which is not known to be free of any of the following harmful organisms:
 - (a) Apple proliferation mycoplasma; or
 - (b) Cherry rasp leaf virus (American),the testing shall use appropriate laboratory methods and, where appropriate, indicator plants for the detection of the relevant harmful organisms, and
- (ii) irrespective of the country of origin of the plant material, the testing shall use appropriate laboratory methods and, where appropriate, indicator plants for the detection of at least the following harmful organisms:
 - (a) Tobacco ringspot virus
 - (b) Tomato ringspot virus
 - (c) *Erwinia amylovora* (Burr.) Winsl. *et al.*

3.3. In the case of *Prunus* L., as appropriate for each *Prunus* species:

- (i) where the plant material originates from a country which is not known to be free of any of the following harmful organisms:
 - (a) Apricot chlorotic leafroll mycoplasma;
 - (b) Cherry rasp leaf virus (American); or
 - (c) *Pseudomonas syringae* pv. *persicae* (Prunier *et al.*) Young *et al.*,the testing shall use appropriate laboratory methods and, where appropriate, indicator plants for the detection of the relevant harmful organisms; and
- (ii) irrespective of the country of origin of the plant material, the testing shall use appropriate laboratory methods and, where appropriate, indicator plants for the detection of at least the following harmful organisms:
 - (a) Little cherry pathogen (non-European isolates)
 - (b) Peach mosaic virus (American)
 - (c) Peach phony rickettsia
 - (d) Peach rosette mosaic virus
 - (e) Peach rosette mycoplasma

- (f) Peach X-disease mycoplasma
 - (g) Peach yellows mycoplasma
 - (h) Plum line pattern virus (American)
 - (i) Plum pox virus
 - (j) Tomato ringspot virus
 - (k) *Xanthomonas campestris* pv. *pruni* (Smith) Dye.
- 3.4. In the case of *Cydonia* Mill. and *Pyrus* L. irrespective of the country of origin of the plant material, testing by appropriate laboratory methods, and, where appropriate, indicator plants, for detection of at least the following harmful organisms:
- (a) *Erwinia amylovora* (Burr.) Winsl. *et al.*
 - (b) Pear decline mycoplasma.
4. The plant material subjected to the visual inspections referred to in point 2 and on which signs and symptoms of harmful organisms have been observed shall be subject to an investigation including testing where necessary, to determine, as far as possible, the identity of the harmful organisms causing the signs and symptoms.

Section III:

Plants of *Vitis* L., other than fruits

1. The plant material shall be subjected, as appropriate, to appropriate therapy procedures, as laid down in FAO/IPGRI Technical Guidelines.
2. The plant material, following the therapy procedures carried out pursuant to point 1, shall be subjected to indexing procedures in its entirety. All plant material including indexing plants, shall be held at the approved facilities under the quarantine containment conditions laid down in Annex I. Plant material intended for approval for official release shall be held under conditions conducive to a normal cycle of vegetative growth and shall be subjected to visual inspection for signs and symptoms of harmful organisms including those of *Daktulosphaira vitifoliae* (Fitch) and of all other relevant harmful organisms listed in Directive 2000/29/EC, on arrival and subsequently, at appropriate times, during the period of the indexing procedures.
3. For the purposes of point 2 the plant material shall be indexed for harmful organisms (tested for and identified) according to the following procedures:
 - 3.1. Where the plant material originates in a country which is not known to be free of the following harmful organisms:
 - (i) *Ajinashika disease*
The testing shall use an appropriate laboratory method. In the event of a negative result, the plant material shall be indexed on the vine variety Koshu and kept under observation during at least two cycles of vegetation.
 - (ii) *Grapevine stunt virus*

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The testing shall use appropriate indicator plants, including the vine variety Campbell Early, and observation shall take place during one year.

(iii) *Summer mottle*

The testing shall use appropriate indicator plants, including the vine varieties Sideritis, Cabernet-Franc and Mission.

3.2. Irrespective of the country of origin of the plant material, the testing shall use appropriate laboratory methods and, where appropriate, indicator plants for the detection of at least the following harmful organisms:

- (a) Blueberry leaf mottle virus
- (b) Grapevine Flavescence dorée MLO and other grapevine yellows
- (c) Peach rosette mosaic virus
- (d) Tobacco ringspot virus
- (e) Tomato ringspot virus (strain 'yellow vein' and other strains)
- (f) *Xylella fastidiosa* (Well & Raju)
- (g) *Xylophilus ampelinus* (Panagopoulos) Willems *et al.*

4. The plant material subjected to the visual inspections referred to in point 2 and on which signs and symptoms of harmful organisms have been observed shall be subjected to an investigation including testing where necessary, to determine, as far as possible, the identity of the harmful organisms causing the signs and symptoms.

Section IV:

Plants of stolon- or tuber-forming species of *Solanum L. or their hybrids, intended for planting*

1. The plant material, as appropriate, shall be subjected to the therapy procedures as laid down in FAO/IPGRI Technical Guidelines.
2. Each unit of the plant material, following the therapy procedures carried out pursuant to point 1, shall be subjected to indexing procedures. All plant material, including indexing plants, shall be held at the approved facilities under the quarantine containment conditions laid down in Annex I. Plant material intended for approval for official release shall be held under conditions conducive to a normal cycle of vegetative growth and be subjected to visual inspection for signs and symptoms of harmful organisms including all relevant harmful organisms listed in Directive 2000/29/EC and potato yellow vein disease, on arrival and subsequently, at regular intervals until senescence, during the period of the indexing procedures.
3. The indexing procedures referred to in point 2 shall follow the technical provisions set out in point 5, in order to detect at least the following harmful organisms:
 - *Bacteria*
 - (a) *Clavibacter michiganensis* (Smith) Davis *et al.* ssp. *sepedonicus* (Speckermann et Kotthoff) Davis *et al.*;

- (b) *Ralstonia solanacearum* (Smith) Yabuuchi *et al.*
- *Viruses and virus-like organisms*
 - (a) Andean potato latent virus,
 - (b) Potato black ringspot virus,
 - (c) Potato spindle tuber viroid,
 - (d) Potato yellowing alfamovirus,
 - (e) Potato virus T,
 - (f) Andean potato mottle virus,
 - (g) Common potato viruses A, M, S, V, X and Y (including Y^o, Yⁿ and Y^c) and potato leaf roll virus.

However, in the case of true seed of potato, the indexing procedures shall be carried out in order to detect at least the viruses and virus-like organisms listed above at (a) to (e).

4. The plant material subjected to the visual inspections referred to in point 2 and on which signs and symptoms of harmful organisms have been observed shall be subjected to an investigation including testing where necessary, to determine, as far as possible, the identity of the harmful organisms causing the signs and symptoms.

5. The technical provisions referred to in point 3 shall be as follows:

- *Bacteria*

1. For tubers, test the heel end of each tuber. The standard sample size shall be 200 tubers. However, the procedure can be applied for samples with less than 200 tubers.
2. For young plants and cuttings, including micro-plants, test the lower sections of the stem and, where appropriate, the roots, for each unit of the plant material.
3. For testing of progeny tubers, or of stem bases for non-tuber forming species, one normal cycle of vegetative growth after the testing referred to in points 1 and 2, is recommended.
4. For the material referred to in point 1, the testing method for *Clavibacter michiganensis* (Smith) Davis *et al.* ssp. *sepedonicus* (Spieckermann et Kotthoff) Davis *et al.* shall be the Community method set out in Annex I to Council Directive 93/85/EEC⁽¹⁾. For the material referred to in point 2, this testing method could be applied.
5. For the material referred to in point 1, the testing method for *Ralstonia solanacearum* (Smith) Yabuuchi *et al.* shall be the test scheme set out in Annex II to Council Directive 98/57/EC⁽²⁾. For the material referred to in point 2, this testing method could be applied.

- *Viruses and virus-like organisms, other than potato spindle tuber viroid*

1. The minimum testing for vegetative material (tubers, young plants and cuttings, including micro-plants) shall include a serological test done at or near flowering for each of the specified list of harmful organisms other than

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potato spindle tuber viroid, and followed by a biological test of material testing negative in the serological test. For Potato leaf roll virus, two serological tests shall be done.

2. The minimum testing for true seed shall be a serological test or a biological test if no serological test is available. Retesting of a proportion of negative samples and testing of borderline results by another method is highly recommended.
3. The serological and biological testing referred to in points 1 and 2 shall be done on glasshouse grown plants, sampled from at least two positions on every stem, including a young fully expanded leaflet at the top of each stem and an older leaflet from a midway position; each stem shall be sampled because of possible non-systemic infection. In the case of the serological testing, no bulking of leaflets from separate plants shall be done, unless the bulking rate has been validated for the method of use; leaflets from each stem may however be bulked to make up the sample from each plant. In the case of the biological testing, the maximum bulking is up to five plants with inoculation of a minimum of duplicate indicator plants.
4. The appropriate indicator plants to be used for the biological testing referred to in points 1 and 2 shall be those listed by the European and Mediterranean Plant Protection Organization (EPPO), or other officially approved indicator plants, which have been shown to detect the viruses.
5. Only material which has been directly tested shall be released from quarantine. Where eye indexing has been done, only the progeny of the tested eye may be released. The tuber should not be released because of possible problems with non-systemic infection.

— *Potato spindle tuber viroid*

1. For all material, glasshouse grown plants shall be tested, as soon as they are well established but prior to flowering and pollen production. Testing on tuber sprouts/*in vitro* plants/small seedlings shall only be regarded as a preliminary test.
2. Samples shall be taken from a fully expanded leaflet at the top of each stem of the plant.
3. All material for testing shall be grown at temperatures not less than 18 °C (preferably at temperatures higher than 20 °C) and with at least a 16-hour photo-period.
4. Testing shall be by radioactive or non-radioactive labelled cDNA or RNA-probes, return-PAGE (with silver staining) or RT-PCR.
5. The maximum bulking rate for probes and return-PAGE is 5. Use of this or higher bulking rates must be validated.

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- (1) OJ L 259, 18.10.1993, p. 1. Directive as amended by Commission Directive 2006/56/EC (OJ L 182, 4.7.2006, p. 1).
- (2) OJ L 235, 21.8.1998, p. 1. Directive as amended by Commission Directive 2006/63/EC (OJ L 206, 27.7.2006, p. 36).