

**Schemes for the production of healthy plants for planting**  
**Schémas pour la production de végétaux sains destinés à la plantation**

## **Production of pathogen-tested herbaceous ornamentals**

### **Specific scope**

This standard describes the production of pathogen-tested material of herbaceous ornamental plants produced in glasshouse.

### **Specific approval and amendment**

First approved in 2007-09.

This standard initially presents a generalized description of the performance of a propagation scheme for the production of pathogen tested plants and then, in the appendices, presents details of the ornamental plants for which it can be used together with lists of pathogens of concern and recommended test methods. The performance of this scheme follows the general sequence proposed by the EPPO Panel on Certification of Pathogen-tested Ornamentals and adopted by EPPO Council (OEPP/EPPO, 1991). According to this sequence, all plant material that is finally sold derives from an individual nuclear stock plant that has been carefully selected and rigorously tested to ensure the highest practical health status; thereafter, the nuclear stock plants and the propagation stock plants derived from it are maintained under controlled conditions in glasshouses to limit the risk of any infection.

### **1. Selection of candidate plants**

The candidate material may be new cultivars, good-quality material of existing cultivars, tissue cultures or meristem-tip cultures of any of these. Material originating from outside the EPPO region should also be inspected for all EPPO-listed pests<sup>1</sup> and tested under quarantine for all EPPO-listed pests of the crop concerned occurring in the region of origin, and generally inspected for any other pests. Inspections and tests should be performed according to the relevant EPPO phytosanitary procedures.

<sup>1</sup>EPPO-listed pests are pests listed in EPPO Standard PM 1/2 *EPPO A1 and A2 Lists of pests recommended for regulation as quarantine pests*.

### **2. Maintenance and testing of candidate plants for nuclear stock**

#### **2.1 Growing conditions**

The candidate plants for nuclear stock should be kept 'in quarantine', that is, in an isolated, suitably designed, aphid-proof house, separately from the nuclear stock and other material, where it can be observed and tested. All plants should be grown in individual pots containing new or sterilized growing medium that are physically separated from each other to prevent any direct contact between plants, with precautions against infection by pests.

#### **2.2 Testing requirements**

All candidate plants should be individually tested for the major and minor pathogens specified in the appropriate Appendix for the plant concerned where recommended test methods for these pathogens are given.

In general testing of plants on indicator plants is recommended because this will act as a general screen for viruses that can be transmitted by sap (mechanically). In addition, when candidate plants may have been in contact with soil or soil water (e.g. originate from open-field cultivation), they should also be screened for Nepoviruses and Tobraviruses (using either *Nicotiana occidentalis* or *N. benthamina*, and *Chenopodium quinoa*).

The plants should be visually inspected regularly for all pests. When plants are grown in soil, the soil should be free of vector nematodes. Apart from the vector pests which can be adequately controlled (e.g. aphids, *Frankliniella occidentalis* and *Bemisia tabaci*), any plant found to be infected, by testing or by visual examination, should be immediately eliminated.

### 2.3 Promotion to nuclear stock

The plants that give negative results in all tests and inspections can be used to produce nuclear stock plants; depending on the crop, this can be by direct transfer of the tested plant to nuclear stock conditions or by propagation by tissue culture, cuttings, bulbs, tubers etc. Promotion to nuclear stock and transfer to the nuclear stock conditions can only occur following verification and authorization that all required tests and observations have been performed with negative results. This scheme may be used to establish an official certification scheme. In case of an official certification scheme the certifying authority monitors this process.

## 3. Maintenance and testing of the nuclear stock

### 3.1 Growing conditions

For some crops, the nuclear stock can be maintained *in vitro* and, in this form, the clones derived from these individual plants will retain the same status in the scheme. Otherwise, *in vivo* nuclear-stock plants should be kept in a suitably designed aphid-proof house, containing only nuclear-stock plants. They should be maintained under the same conditions, and with the same precautions against infection as candidate nuclear-stock plants (see point 2 above). A check on trueness to type should be made; for many ornamental crops this is done by bringing either the nuclear stock plants, or cuttings/bulbs taken from them, to flower, but the flowering may need to be done in a different place to avoid risk of infection.

The useful life of a nuclear-stock plant is generally limited by the longevity of the individual plants of the species. In other cases, because of the risk of re-infection, the *in vivo* nuclear stock plants should be re-tested after the period of time as specified in the appropriate Appendix for the plant concerned. In addition, when maintenance is done *in vitro*, retesting is recommended.

### 3.2 Testing requirements

All plants should be individually tested for the pathogens specified in the appropriate Appendix for the plant concerned at least once during the productive life of the plant. The frequency of testing should reflect the risk of infection and how long the nuclear stock plants are kept.

Appropriate control measures should be applied to control the other pests specified in the Appendix for the plant concerned. The plants should be visually inspected at regular intervals for all pests. Any plant showing infestation should be eliminated.

Plants propagated from nuclear-stock plants can also be considered as nuclear stock, provided that they do not leave the nuclear stock conditions<sup>2</sup> and are individually tested as above.

<sup>2</sup>They may be transferred to other, similar, nuclear stock conditions and still retain nuclear stock status provided that they are transported while packed at all times in suitable containers designed to avoid contamination (e.g. aphid-proof).

The same applies to plants transferred from *in vitro* culture to pots. In general, a regular control of trueness to type is also necessary when maintaining material *in vivo* or *in vitro*.

### 3.3 Authorization

Before a nuclear stock plant may be propagated further in the scheme, the passage of propagating material to the next stage should be authorized on the basis of records of the tests and observations performed during production, and of one or more authorization (visual) inspections. At inspection, the nuclear stock plant should show no symptoms of fungal, bacterial and viral diseases (unless they can be adequately controlled), and should be free of vector insects (e.g. aphids or thrips), otherwise the plant should be refused. If propagating material from nuclear stock leaves the scheme, it can be referred to as 'pre-basic' material (provided this has been officially authorized).

## 4. Propagation stock I

### 4.1 Growing conditions

Cuttings, or other plant parts used for propagation depending on the crop, taken from the nuclear-stock plants when planted become propagation stock I.

The plants should be kept in isolated houses, separate from any other plants that are not at an equivalent stage of a similar scheme. Plants should be grown either in individual containers or in a system of small growing units ensuring adequate isolation. General precautions against pests should be maintained.

The number of generations of propagation stock I that should not be exceeded and the useful life of a propagation stock I plant are indicated in the Appendix for the plant concerned. After this period all the propagation-stock I plants should either be retested or replaced by new plants. The filiation of the plants should be recorded, so that each lot is known to be derived from nuclear stock by not more than the fixed number of generations of propagation under the required conditions.

Throughout the production of propagation stock I, checks should be made on varietal purity and on possible mutations or back mutations.

### 4.2 Testing requirements

The minimum requirements for Propagation stock I plants are given in the Appendix for the plant concerned together with the recommended tests methods for these pathogens. At this stage of the scheme, the testing requirement is commonly random testing of lots or (sub-lots) of propagation-stock I plants, or visual inspection for specific symptoms. Any plant giving a positive result at random testing should be eliminated and recorded. In the case of a positive test result, all plants in the group of plants from which the sample was taken (whole lot or sub-unit) should be tested individually. All positive plants should be eliminated. The plants should be visually inspected regularly for the presence of any pest. Any plant found to be

substantially infested by any pest should be eliminated, except in the case of pests which can be adequately controlled.

#### 4.3 Authorization

Authorization for further propagation will be granted on the basis of records of the tests and observations performed during production and of one or more (visual) inspections. At final inspection, the plants should be completely free of symptoms of the pests listed in the Appendix for the crop concerned. They should be substantially free of other pests. If these conditions are not met, the lots concerned should not be used for further propagation. Other specific standards may be recommended in the Appendix for the plant concerned. If propagating material from propagation stock I leaves the scheme, it can be referred to as 'basic' material (provided this has been officially authorized).

### 5. Propagation stock II

#### 5.1 Growing conditions

Cuttings, or other plant parts used for propagation depending on the crop, taken from the propagation stock I plants, or plants maintained at nuclear stock, when planted, become the propagation stock II, from which later on the certified material for sale is taken. The propagation stock II plants may be grown in the field or under protection (glasshouses, gauze houses, etc). In general, the plants should be isolated from other plants that are not at an equivalent stage of a similar scheme. If there are any other requirements for the growing conditions, they will

be given in the Appendix for the plant concerned. General precautions against pests should be maintained. An effective crop protection program should be in place.

The number of generations of propagation stock II that should not be exceeded and the useful life of a propagation stock II plant are indicated for each crop in the relevant appendix. After this period all the propagation-stock II plants should be either retested or replaced by new plants. Throughout the production of propagation stock II, checks should be made on varietal purity and on possible mutations or back mutations.

#### 5.2 Testing requirements

The minimum requirements for Propagation stock II plants are given in the Appendix for the plant concerned. At this stage of the scheme, the requirement is most commonly visual inspection for specific symptoms or, occasionally, random testing of lots or (sub-lots) of propagation stock II plants. The plants should be visually inspected regularly for the presence of any pest. Any plant found to be substantially infested by any pest should be eliminated.

#### 5.3 Authorization

Authorization for further propagation will be granted on the basis of records of the tests and observations performed during production and of one or more (visual) inspections. Recommended standards are given in the Appendix for the crop concerned. Propagation material from propagation stock II leaving the scheme can be referred to 'certified' material (if this has been officially authorized).

## Appendix 1

### Alstroemeria

#### Species covered

*Alstroemeria hybrids*

#### Propagation

Dividing, tissue culture, seed

#### Pathogens

Genus	Species	Acronym	Status*	Test method	Vector
<i>Potyvirus</i>	<i>Alstroemeria flower banding virus</i>	AIFBV	Minor	ELISA	Aphids
<i>Potyvirus</i>	<i>Alstroemeria mosaic virus</i>	AIMV	Major	Indicator plants, ELISA	<i>Myzus persicae</i>
<i>Cucumovirus</i>	<i>Cucumber mosaic virus</i>	CMV	Major	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	Aphids
<i>Tospovirus</i>	<i>Impatiens necrotic spot virus</i>	INSV	Major	Indicator plants ( <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA See EPPO Diagnostic Protocol PM 7/34	<i>F. occidentalis</i> , <i>Frankliniella fusca</i>
<i>Tospovirus</i>	<i>Iris yellow spot virus</i>	IYSV	Minor	Indicator plants ( <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA	<i>Thrips tabaci</i>
<i>Potyvirus</i>	<i>Lily mottle virus</i>	LMoV	Minor	ELISA	Aphids
<i>Carlavirus</i>	<i>Lily symptomless virus</i>	LSV	Major	ELISA	<i>Myzus persicae</i>
<i>Potyvirus</i>	<i>Ornithogalum mosaic virus</i>	OrMV	Minor	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	Aphids
<i>Tobravirus</i>	<i>Tobacco rattle virus</i>	TRV	Minor	Indicator plants (e.g. <i>C. quinoa</i> ), PCR†, ELISA‡	<i>Paratrichodorus</i> spp. and <i>Trichodorus</i> spp.
<i>Tospovirus</i>	<i>Tomato spotted wilt virus</i>	TSWV	Major	Indicator plants ( <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA See EPPO Diagnostic Protocol PM 7/34	<i>F. occidentalis</i> , <i>Thrips tabaci</i> , <i>T. setosus</i> , <i>F. fusca</i> , <i>F. intonsa</i> , <i>F. schultzei</i> , <i>Scirtothrips dorsalis</i>

\*Pests categorized as major pests should be tested/checked/monitored during the entire propagation scheme, whereas it is recommended that the 'minor pests' are checked (if necessary with testing) at least during the selection and testing of the candidate nuclear stock plants.

†Cornelissen BJC, Linthorst HJM, Brederode FT & Bol JF (1986) Analysis of the genome structure of tobacco rattle virus strain PSG. *Nucleic Acids Research* **14**(5), 2157–2169.

‡Several sets of antisera should be used to detect various serotypes of the virus.

### Special requirements in the scheme

*Useful life of Nuclear Stock Plant*

1 year

*Maximum number of generations of Propagation Stock*

Propagation Stock I: 2

Propagation Stock II: 1

*Pests especially requiring control*

*Liriomyza* spp. and *Pratylenchus* spp.

## Appendix 2

### Anthurium

#### Species covered

*Anthurium* spp. such as *A. andraeanum* and its hybrids

#### Propagation

Tissue culture, cuttings, seed

#### Pathogens

Genus	Species	Acronym	Status*	Test method	Vector
<i>Cucumovirus</i>	<i>Cucumber mosaic virus</i>	CMV	Major	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	Aphids
<i>Potyvirus</i>	<i>Dasheen mosaic virus</i>	DsMV	Major	Indicator plant ( <i>Philodendron selloum</i> ), ELISA	Aphids
<i>Tospovirus</i>	<i>Impatiens necrotic spot virus</i>	INSV	Major	Indicator plants ( <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA See EPPO Diagnostic Protocol PM 7/34	<i>F. occidentalis</i> , <i>Frankliniella fusca</i>
<i>Potyvirus</i>	<i>Konjac mosaic virus</i>	KoMV	Major	ELISA	Aphids
<i>Tospovirus</i>	<i>Tomato spotted wilt virus</i>	TSWV	Major	Indicator plants ( <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA See EPPO Diagnostic Protocol PM 7/34	<i>F. occidentalis</i> , <i>Thrips tabaci</i> , <i>T. setosus</i> , <i>F. fusca</i> , <i>F. intonsa</i> , <i>F. schultzei</i> , <i>Scirtothrips dorsalis</i>
<i>Xanthomonas</i>	<i>Xanthomonas axonopodis</i> pv. <i>dieffenbachiae</i>	–	Major	Culturing, IF, ELISA See EPPO Diagnostic Protocol PM 7/23	–

\*Pests categorized as major pests should be tested/checked/monitored during the entire propagation scheme, whereas it is recommended that the 'minor pests' are checked (if necessary with testing) at least during the selection and testing of the candidate nuclear stock plants.

#### Special requirements in the scheme

*Useful life of Nuclear Stock Plant*

1 year

*Maximum number of generations of Propagation Stock*

Propagation Stock I: 2

Propagation Stock II: 1

*Pests especially requiring control*

*Radopholus similis*

## Appendix 3

### Argyranthemum Frutescens

#### Species covered

*Argyranthemum frutescens*

#### Propagation

Cuttings, tissue culture, seed

#### Pathogens

Genus	Species	Acronym	Status*	Test method	Vector
<i>Luteovirus</i>	<i>Beet western yellows virus</i>	BWYV	Minor	ELISA	Aphids
<i>Pospiviroid</i>	<i>Chrysanthemum stunt viroid</i>	CSVd	Major	R-PAGE, Nucleic acid hybridization, RT-PCR, Fluorogenic 5-nuclease assay (TaqMan) See EPPO Diagnostic Protocol PM 7/6	None, spread by plant sap
<i>Carlavirus</i>	<i>Chrysanthemum virus B</i>	CVB	Major	ELISA	Aphids
<i>Carlavirus</i>	<i>Helenium virus S</i>	HVS	Minor	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	<i>Myzus persicae</i>
<i>Tospovirus</i>	<i>Impatiens necrotic spot virus</i>	INSV	Major	Indicator plants ( <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA See EPPO Diagnostic Protocol PM 7/34	<i>F. occidentalis</i> , <i>Frankliniella fusca</i>
<i>Potyvirus</i>	Potyviruses		Minor	Broad-spectrum ELISA, PCR†	Aphids
<i>Cucumovirus</i>	<i>Tomato aspermy virus</i>	TAV	Minor	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	Aphids
<i>Tospovirus</i>	<i>Tomato spotted wilt virus</i>	TSWV	Major	Indicator plants ( <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA See EPPO Diagnostic Protocol PM 7/34	<i>F. occidentalis</i> , <i>Thrips tabaci</i> , <i>T. setosus</i> , <i>F. fusca</i> , <i>F. intonsa</i> , <i>F. schultzei</i> , <i>Scirtothrips dorsalis</i>
<i>Agrobacterium</i>	<i>Agrobacterium tumefaciens</i>	–	Major	Culturing and then confirmation by PCR	–
<i>Dickeya</i>	<i>Dickeya (Erwinia) chrysanthemi</i>	–	Minor	Culturing and then confirmation by fatty acids analysis IF for screening	–
<i>Pseudomonas</i>	<i>Pseudomonas cichorii</i>	–	Minor	Culturing and then confirmation by fatty acids analysis	–
<i>Phytophthora</i>	<i>Phytophthora tentaculata</i>	–	Minor	Culturing, morphological identification	–
	rust‡	–	Minor		–
<i>Peronospora</i>	<i>Peronospora radii</i>		Minor		

\*Pests categorized as major pests should be tested/checked/monitored during the entire propagation scheme, whereas it is recommended that the 'minor pests' are checked (if necessary with testing) at least during the selection and testing of the candidate nuclear stock plants.

†e.g. Langeveld SA, Dore JM, Memelink J, Derks AFLM, Vlugt CIM van der, Asjes CJ & Bol JF (1991) Identification of potyviruses using the polymerase chain reaction with degenerate primers. *Journal of General Virology* **72**, 1531–1541.

‡This species can not be identified; check should be done by visual inspection.

#### Special requirements in the scheme

*Useful life of Nuclear Stock Plant*

1 year

*Maximum number of generations of Propagation Stock*

Propagation Stock I: 2

Propagation Stock II: 1

*Pests especially requiring control*

*Aphelenchoides* spp.

*Liriomyza* spp.

## Appendix 4

### Brachyscome

#### Species covered

*Brachyscome* spp. such as *B. iberidifolia*

#### Propagation

Cuttings, tissue culture, seed

#### Main pests

##### Viruses

Genus	Species	Acronym	Status*	Test method	Vector
<i>Cucumovirus</i>	<i>Cucumber mosaic virus</i>	CMV	Major	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	Aphids
<i>Tospovirus</i>	<i>Impatiens necrotic spot virus</i>	INSV	Major	Indicator plants ( <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA See EPPO Diagnostic Protocol PM 7/34	<i>F. occidentalis</i> , <i>Frankliniella fusca</i>
<i>Potyvirus</i>	Potyviruses		Major	Broad-spectrum ELISA, PCR†	Aphids
<i>Tospovirus</i>	<i>Tomato spotted wilt virus</i>	TSWV	Major	Indicator plants ( <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA See EPPO Diagnostic Protocol PM 7/34	<i>F. occidentalis</i> , <i>Thrips tabaci</i> , <i>T. setosus</i> , <i>F. fusca</i> , <i>F. intonsa</i> , <i>F. schultzei</i> , <i>Scirtothrips dorsalis</i>

\*Pests categorized as major pests should be tested/checked/monitored during the entire propagation scheme, whereas it is recommended that the 'minor pests' are checked (if necessary with testing) at least during the selection and testing of the candidate nuclear stock plants.

†e.g. Langeveld SA, Dore JM, Memelink J, Derks AFLM, Vlugt CIM van der, Asjes CJ & Bol JF (1991) Identification of potyviruses using the polymerase chain reaction with degenerate primers. *Journal of General Virology* **72**, 1531–1541.

#### Special requirements in the scheme

##### Useful life of Nuclear Stock Plant

1 year

##### Maximum number of generations of Propagation Stock

Propagation Stock I: 2

Propagation Stock II: 1

## Appendix 5

### Bracteantha

#### Species covered

*Bracteantha* spp. such as *B. bracteata*, syn. *Helichrysum bracteatum*, syn. *Xerochrysum bracteatum*

#### Propagation

Cuttings, tissue culture, seed

#### Pathogens

Genus	Species	Acronym	Status*	Test method	Vector
<i>Carlavirus</i>	<i>Chrysanthemum virus B</i>	CVB	Major	ELISA	Aphids
<i>Potyvirus</i>	Potyriviruses		Major	Broad-spectrum ELISA, PCR†	Aphids
<i>Cucumovirus</i>	<i>Tomato aspermy virus</i>	TAV	Major	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	Aphids
<i>Dickeya</i>	<i>Dickeya (Erwinia) chrysanthemi</i>	–	Minor	Culturing and then confirmation by fatty acids analysis IF for screening	–

\*Pests categorized as major pests should be tested/checked/monitored during the entire propagation scheme, whereas it is recommended that the 'minor pests' are checked (if necessary with testing) at least during the selection and testing of the candidate nuclear stock plants.

†e.g. Langeveld SA, Dore JM, Memelink J, Derks AFLM, Vlugt CIM van der, Asjes CJ & Bol JF (1991) Identification of potyriviruses using the polymerase chain reaction with degenerate primers. *Journal of General Virology* **72**, 1531–1541.

## ELISA

### Special requirements in the scheme

*Useful life of Nuclear Stock Plant*

1 year

*Maximum number of generations of Propagation Stock*

Propagation Stock I: 2

Propagation Stock II: 1



## Appendix 6

### Campanula

#### Species covered

*Campanula* spp. and hybrids

#### Propagation

Seed, cuttings, tissue culture

#### Pathogens

Genus	Species	Acronym	Status*	Test method	Vector
<i>Cucumovirus</i>	<i>Cucumber mosaic virus</i>	CMV	Major	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	Aphids
<i>Tospovirus</i>	<i>Impatiens necrotic spot virus</i>	INSV	Major	Indicator plants ( <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA See EPPO Diagnostic Protocol PM 7/34	<i>F. occidentalis</i> , <i>Frankliniella fusca</i>
<i>Potyvirus</i>	Potyviruses		Major	Broad-spectrum ELISA, PCR†	Aphids
<i>Nepovirus</i>	<i>Tomato black ring virus</i>	TBRV	Minor	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	<i>Longidorus elongatus</i> and <i>L. attenuatus</i>
<i>Tospovirus</i>	<i>Tomato spotted wilt virus</i>	TSWV	Major	Indicator plants ( <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA See EPPO Diagnostic Protocol PM 7/34	<i>F. occidentalis</i> , <i>Thrips tabaci</i> , <i>T. setosus</i> , <i>F. fusca</i> , <i>F. intonsa</i> , <i>F. schultzei</i> , <i>Scirtothrips dorsalis</i>
<i>Plectosporium</i>	<i>Plectosporium tabacinum</i>	–	Minor	Culturing, morphological identification	–

\*Pests categorized as major pests should be tested/checked/monitored during the entire propagation scheme, whereas it is recommended that the ‘minor pests’ are checked (if necessary with testing) at least during the selection and testing of the candidate nuclear stock plants.

†e.g. Langeveld SA, Dore JM, Memelink J, Derks AFLM, Vlugt CIM van der, Asjes CJ & Bol JF (1991) Identification of potyviruses using the polymerase chain reaction with degenerate primers. *Journal of General Virology* **72**, 1531–1541.

#### Special requirements in the scheme

Propagation Stock II: 1

#### Useful life of Nuclear Stock Plant

1 year

#### Maximum number of generations of Propagation Stock

Propagation Stock I: 2

## Appendix 7

### Dahlia

#### Species covered

*Dahlia hybrids*

#### Propagation

Cuttings, tissue culture, dividing, seed

#### Pathogens

Genus	Species	Acronym	Status*	Test method	Vector
<i>Alfavirus</i>	<i>Alfalfa mosaic virus</i>	AMV	Minor	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	Aphids
<i>Cucumovirus</i>	<i>Cucumber mosaic virus</i>	CMV	Major	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	Aphids
<i>Caulimovirus</i>	<i>Dahlia mosaic virus</i>	DMV	Major	ELISA, PCR†	Aphids
<i>Tospovirus</i>	<i>Impatiens necrotic spot virus</i>	INSV	Major	Indicator plants ( <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA See EPPO Diagnostic Protocol PM 7/34	<i>F. occidentalis</i> , <i>Frankliniella fusca</i>
<i>Iarvirus</i>	<i>Tobacco streak virus</i>	TSV	Major	Indicator plants <i>C. quinoa</i> and <i>N. occidentalis</i> , ELISA	Pollen transported by thrips
<i>Tospovirus</i>	<i>Tomato spotted wilt virus</i>	TSWV	Major	Indicator plants ( <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA See EPPO Diagnostic Protocol PM 7/34	<i>F. occidentalis</i> , <i>Thrips tabaci</i> , <i>T. setosus</i> , <i>F. fusca</i> , <i>F. intonsa</i> , <i>F. schultzei</i> , <i>Scirtothrips dorsalis</i>
<i>Dickeya</i>	<i>Dickeya (Erwinia) chrysanthemi</i>	–	Minor	Culturing and then confirmation by fatty acids analysis IF for screening	–
<i>Agrobacterium</i>	<i>Agrobacterium tumefaciens</i>	–	Minor	Culturing and then confirmation by PCR	–
<i>Rhodococcus</i>	<i>Rhodococcus fascians</i>	–	Minor	Culturing and then confirmation by PCR‡	–
<i>Verticillium</i>	<i>Verticillium albo-atrum</i>	–	Minor	Culturing, morphological identification	–

\*Pests categorized as major pests should be tested/checked/monitored during the entire propagation scheme, whereas it is recommended that the ‘minor pests’ are checked (if necessary with testing) at least during the selection and testing of the candidate nuclear stock plants.

†Nicolaisen M (2003) Partial molecular characterization of Dahlia mosaic virus and its detection by PCR. *Plant Disease* **87**, 945–948.

‡Stange RR Jr, Jeffares D, Young C, Scott DB, Eason JR & Jameson PE (1996) PCR amplification of the *fas-1* gene for the detection of virulent strains of *Rhodococcus fascians*. *Plant Pathology* **45**, 407–417.

#### Special requirements in the scheme

*Useful life of Nuclear Stock Plant*

1 year

Propagation Stock I: 2

Propagation Stock II: 1

*Maximum number of generations of Propagation Stock*

## Appendix 8

### Fuchsia

#### Species covered

*Fuchsia* spp. and hybrids

#### Propagation

Cutting, tissue culture, seed

#### Pathogens

Genus	Species	Acronym	Status*	Test method	Vector
<i>Nepovirus</i>	Arabis mosaic virus	ArMV	Minor	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	<i>Xiphinema</i> spp.
<i>Cucumovirus</i>	Cucumber mosaic virus	CMV	Major	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	Aphids
<i>Tospovirus</i>	<i>Impatiens necrotic spot</i>	INSV	Major	Indicator plants ( <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA See EPPO Diagnostic Protocol PM 7/34	<i>F. occidentalis</i> , <i>Frankliniella fusca</i>
<i>Tospovirus</i>	Tomato spotted wilt virus	TSWV	Major	Indicator plants ( <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA See EPPO Diagnostic Protocol PM 7/34	<i>F. occidentalis</i> , <i>Thrips tabaci</i> , <i>T. setosus</i> , <i>F. fusca</i> , <i>F. intonsa</i> , <i>F. schultzei</i> , <i>Scirtothrips dorsalis</i>

\*Pests categorized as major pests should be tested/checked/monitored during the entire propagation scheme, whereas it is recommended that the 'minor pests' are checked (if necessary with testing) at least during the selection and testing of the candidate nuclear stock plants.

NB: *Fuchsia latent virus* is also a pest of concern but further information is needed.

#### Special requirements in the scheme

Useful life of Nuclear Stock Plant

1 year

Maximum number of generations of Propagation Stock

Propagation Stock I: 2

Propagation Stock II: 1

## Appendix 9

### Gentiana

#### Species covered

*Gentiana* spp. and hybrids

#### Propagation

Cuttings, tissue culture, dividing, seed

#### Pathogens

Genus	Species	Acronym	Status*	Test method	Vector
<i>Potyvirus</i>	Bean yellow mosaic virus	BYMV	Major	Indicator plants (both <i>C. quinoa</i> and <i>Nicotiana occidentalis</i> to detect all strains), ELISA	Aphids
<i>Fabavirus</i>	Broad bean wilt virus 1	BBWV1	Minor	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	Aphids
<i>Fabavirus</i>	Broad bean wilt virus 2	BBWV2	Major	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	Aphids
<i>Potyvirus</i>	Clover yellow vein virus	CIYVV	Major	ELISA	Aphids
<i>Cucumovirus</i>	Cucumber mosaic virus	CMV	Major	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	Aphids
<i>Tospovirus</i>	<i>Impatiens necrotic spot virus</i>	INSV	Major	Indicator plants ( <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA See EPPO Diagnostic Protocol PM 7/34	<i>F. occidentalis</i> , <i>Frankliniella fusca</i>

\*Pests categorized as major pests should be tested/checked/monitored during the entire propagation scheme, whereas it is recommended that the 'minor pests' are checked (if necessary with testing) at least during the selection and testing of the candidate nuclear stock plants.

NB: Gentian mosaic virus is a new virus of concern for Gentiana. It can be detected by indicator plant such as *C. quinoa* and some others, but not *Nicotiana* spp. Further information is still needed.

#### Special requirements in the scheme

Useful life of Nuclear Stock Plant

1 year

Maximum number of generations of Propagation Stock

Propagation Stock I: 2

Propagation Stock II: 1

## Appendix 10

### Hosta

#### Species covered

*Hosta* spp. and hybrids

#### Propagation

Tissue culture, dividing, cuttings, seed

#### Pathogens

Genus	Species	Acronym	Status*	Test method	Vector
<i>Nepovirus</i>	<i>Arabid mosaic virus</i>	ArMV	Minor	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	<i>Xiphinema</i> spp.
<i>Cucumovirus</i>	<i>Cucumber mosaic virus</i>	CMV	Minor	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	Aphids
<i>Potexvirus</i>	<i>Hosta virus X</i>	HVX	Major	ELISA	None, spread by plant sap
<i>Tospovirus</i>	<i>Impatiens necrotic spot virus</i>	INSV	Major	Indicator plants ( <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA See EPPO Diagnostic Protocol PM 7/34	<i>F. occidentalis</i> , <i>Frankliniella fusca</i>
<i>Tobravirus</i>	<i>Tobacco rattle virus</i>	TRV	Minor	Indicator plants (e.g. <i>C. quinoa</i> ), PCR†, ELISA‡	<i>Trichodorus</i> spp., <i>Paratrichodorus</i> spp.
<i>Nepovirus</i>	<i>Tomato ringspot virus</i>	ToRSV	Minor	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA See EPPO Diagnostic Protocol PM 7/49	<i>Xiphinema americanum sensu lato</i>
<i>Tospovirus</i>	<i>Tomato spotted wilt virus</i>	TSWV	Major	Indicator plants ( <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA See EPPO Diagnostic Protocol PM 7/34	<i>F. occidentalis</i> , <i>Thrips tabaci</i> , <i>T. setosus</i> , <i>F. fusca</i> , <i>F. intonsa</i> , <i>F. schultzei</i> , <i>Scirtothrips dorsalis</i>
<i>Nepovirus</i>	<i>Tobacco ringspot virus</i>	TRSV	Minor	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA See EPPO Diagnostic Protocol PM 7/2	<i>Xiphinema americanum sensu lato</i>

\*Pests categorized as major pests should be tested/checked/monitored during the entire propagation scheme, whereas it is recommended that the 'minor pests' are checked (if necessary with testing) at least during the selection and testing of the candidate nuclear stock plants.

†Cornelissen BJC, Linthorst HJM, Brederode FT & Bol JF (1986) Analysis of the genome structure of tobacco rattle virus strain PSG. *Nucleic Acids Research* **14**(5), 2157–2169.

‡Several sets of antisera should be used to detect various serotypes of the virus.

#### Special requirements in the scheme

*Useful life of Nuclear Stock Plant*

1 year

*Maximum number of generations of Propagation Stock*

Propagation Stock I: 2

Propagation Stock II: 1

## Appendix 11

### Impatiens

#### Species covered

*Impatiens* spp. such as *I. walleriana*

#### Propagation

Cuttings, seed, tissue culture

#### Pathogens

Genus	Species	Acronym	Status*	Test method	Vector
<i>Alfavirus</i>	<i>Alfalfa mosaic virus</i>	AMV	Minor	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	Aphids
<i>Cucumovirus</i>	<i>Cucumber mosaic virus</i>	CMV	Major	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	Aphids
<i>Tospovirus</i>	<i>Impatiens necrotic spot virus</i>	INSV	Major	Indicator plants ( <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA See EPPO Diagnostic Protocol PM 7/34	<i>F. occidentalis</i> , <i>Frankliniella fusca</i>
<i>Potyvirus</i>	Potyviruses	–	Minor	Broad-spectrum ELISA, PCR†	Aphids
<i>Tobamovirus</i>	<i>Ribgrass mosaic virus</i>	RMV	Minor	Indicator plants (e.g. <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA	None, spread by plant sap
<i>Tobamovirus</i>	<i>Tobacco mild green mosaic</i>	TMGMV	Major	Indicator plants (e.g. <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA	None, spread by plant sap
<i>Tobamovirus</i>	<i>Tobacco mosaic virus</i>	TMV	Major	Indicator plants (e.g. <i>Nicotiana glutinosa</i> , <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA	None, spread by plant sap
<i>Illavirus</i>	<i>Tobacco streak virus</i>	TSV	Major	Indicator plants <i>C. quinoa</i> and <i>N. occidentalis</i> , ELISA	Pollen transported by thrips
<i>Tobamovirus</i>	<i>Tomato mosaic virus</i>	ToMV	Major	Indicator plants (e.g. <i>Nicotiana glutinosa</i> , <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA	None, spread by plant sap
<i>Tospovirus</i>	<i>Tomato spotted wilt virus</i>	TSWV	Major	Indicator plants ( <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA See EPPO Diagnostic Protocol PM 7/34	<i>F. occidentalis</i> , <i>Thrips tabaci</i> , <i>T. setosus</i> , <i>F. fusca</i> , <i>F. intonsa</i> , <i>F. schultzei</i> , <i>Scirtothrips dorsalis</i>
<i>Plasmopara</i>	<i>Plasmopara obducens</i>	–	Minor		–

\*Pests categorized as major pests should be tested/checked/monitored during the entire propagation scheme, whereas it is recommended that the 'minor pests' are checked (if necessary with testing) at least during the selection and testing of the candidate nuclear stock plants.

†e.g. Langeveld SA, Dore JM, Memelink J, Derks AFLM, Vlucht CIM van der, Asjes CJ & Bol JF (1991) Identification of potyviruses using the polymerase chain reaction with degenerate primers. *Journal of General Virology* **72**, 1531–1541.

#### Special requirements in the scheme

*Useful life of Nuclear Stock Plant*

1 year

*Maximum number of generations of Propagation Stock*

Propagation Stock I: 2

Propagation Stock II: 1

## Appendix 12

### Lobelia

#### Species covered

*Lobelia* spp. such as *L. erinus*

#### Propagation

Cuttings, tissue culture, dividing, seed

#### Pathogens

##### Viruses

Genus	Species	Acronym	Status*	Test method	Vector
<i>Potexvirus</i>	<i>Alternanthera mosaic virus</i>	AltMV	Minor	Indicator plants (e.g. <i>Chenopodium quinoa</i> ), ELISA (can be detected with <i>Papaya Mosaic Virus</i> antiserum)	None, transmitted by sap
<i>Nepovirus</i>	<i>Arabis mosaic virus</i>	ArMV	Major	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	<i>Xiphinema</i> spp.
<i>Fabavirus</i>	<i>Broad bean wilt virus 1</i>	BBWV1	Minor	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	Aphids
<i>Cucumovirus</i>	<i>Cucumber mosaic virus</i>	CMV	Major	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	Aphids
<i>Tospovirus</i>	<i>Impatiens necrotic spot virus</i>	INSV	Major	Indicator plants ( <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA See EPPO Diagnostic Protocol PM 7/34	<i>F. occidentalis</i> , <i>Frankliniella fusca</i>
<i>Potyvirus</i>	Potyriviruses		Minor	Broad-spectrum ELISA, PCR†	Aphids
<i>Tospovirus</i>	<i>Tomato spotted wilt virus</i>	TSWV	Major	Indicator plants ( <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA See EPPO Diagnostic Protocol PM 7/34	<i>F. occidentalis</i> , <i>Thrips tabaci</i> , <i>T. setosus</i> , <i>F. fusca</i> , <i>F. intonsa</i> , <i>F. schultzei</i> , <i>Scirtothrips dorsalis</i>
<i>Xanthomonas</i>	<i>Xanthomonas campestris</i> strain pathogenic for <i>Lobelia</i>	–	Major	Culturing, IF	–

\*Pests categorized as major pests should be tested/checked/monitored during the entire propagation scheme, whereas it is recommended that the ‘minor pests’ are checked (if necessary with testing) at least during the selection and testing of the candidate nuclear stock plants.

†e.g. Langeveld SA, Dore JM, Memelink J, Derks AFLM, Vlugt CIM van der, Asjes CJ & Bol JF (1991) Identification of potyviruses using the polymerase chain reaction with degenerate primers. *Journal of General Virology* **72**, 1531–1541.

#### Special requirements in the scheme

##### Useful life of Nuclear Stock Plant

1 year

##### Maximum number of generations of Propagation Stock

Propagation Stock I: 2

Propagation Stock II: 1

## Appendix 13

### Orchids

#### Species covered

Species and hybrids of glasshouse *Orchidaceae* especially *Phalaenopsis*, *Cymbidium*, *Oncidium*, *Dendrobium*

#### Propagation

Tissue culture, cuttings, dividing, seed

#### Pathogens

Genus	Species	Acronym	Status*	Test method	Vector
<i>Cucumovirus</i>	<i>Cucumber mosaic virus</i>	CMV	Minor	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	Aphids
<i>Potexvirus</i>	<i>Cymbidium mosaic virus</i>	CymMV	Major	Indicator plants (e.g. <i>C. Amaranticolor</i> ), ELISA	None, spread by plant sap
<i>Tospovirus</i>	<i>Impatiens necrotic spot virus</i>	INSV	Major	Indicator plants ( <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA See EPPO Diagnostic Protocol PM 7/34	<i>F. occidentalis</i> , <i>Frankliniella fusca</i>
<i>Tobamovirus</i>	<i>Odontoglossum ringspot virus</i>	ORSV	Major	Indicator plants (e.g. <i>C. quinoa</i> , <i>N. occidentalis</i> ), ELISA	None, spread by plant sap
<i>Rhabdovirus</i>	<i>Orchid fleck virus</i>	OFV	Minor	PCR†	Mites
<i>Potyvirus</i>	<i>Potyrivuses</i>	–	Minor‡	Broad-spectrum ELISA, PCR§	Aphids
<i>Tospovirus</i>	<i>Tomato spotted wilt virus</i>	TSWV	Major	Indicator plants ( <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA See EPPO Diagnostic Protocol PM 7/34	<i>F. occidentalis</i> , <i>Thrips tabaci</i> , <i>T. setosus</i> , <i>F. fusca</i> , <i>F. intonsa</i> , <i>F. schultzei</i> , <i>Scirtothrips dorsalis</i>
<i>Acidovorax</i>	<i>Acidovorax avenae</i> subsp. <i>cattleyae</i>	–	Minor	Immunofluorescence (IF), culturing, PCR¶	–

\*Pests categorized as major pests should be tested/checked/monitored during the entire propagation scheme, whereas it is recommended that the 'minor pests' are checked (if necessary with testing) at least during the selection and testing of the candidate nuclear stock plants.

†Blanchfield AL, Mackenzie AM, Gibbs A, Kondo H, Tamada T & Wilson CR (2001). Identification of Orchid fleck virus by Reverse Transcriptase-Polymerase Chain Reaction and analysis of isolate relationships. *Journal of Phytopathology* **149**, 713–718.

‡Important for genera in *Orchidaceae* other than *Phalaenopsis* (e.g. *Vanilla*, *Cypripedium*).

§e.g. Langeveld SA, Dore JM, Memelink J, Derks AFLM, Vlugt CIM van der, Asjes CJ & Bol JF (1991) Identification of potyviruses using the polymerase chain reaction with degenerate primers. *Journal of General Virology* **72**, 1531–1541.

¶Schaad NW, Jones JB & Chun W (2003) *Acidovorax avenae*. In *Plant Pathogenic Bacteria*. Third Edition, APS press, St. Paul, Minnesota, USA.

#### Special requirements in the scheme

*Useful life of Nuclear Stock Plant*

1 year

*Maximum number of generations of Propagation Stock*

Propagation Stock I: 2

Propagation Stock II: 1

## Appendix 14

### Osteospermum

#### Species covered

*Osteospermum* spp. such as *O. barbarea*

#### Propagation

Cuttings, tissue culture, seed

#### Pathogens

##### Viruses

Genus	Species	Acronym	Status*	Test method	Vector
<i>Alfavirus</i>	<i>Alfalfa mosaic virus</i>	AMV	Minor	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	Aphids
<i>Potyvirus</i>	<i>Bean yellow mosaic virus</i>	BYMV	Minor	Indicator plants (both <i>C. quinoa</i> and <i>Nicotiana occidentalis</i> to detect all strains), ELISA	Aphids
<i>Luteovirus</i>	<i>Beet western yellows virus</i>	BWYV	Minor	ELISA	Aphids
<i>Carlavirus</i>	<i>Chrysanthemum B virus</i>	CVB	Minor	ELISA	Aphids
<i>Cucumovirus</i>	<i>Cucumber mosaic virus</i>	CMV	Major	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	Aphids
<i>Tospovirus</i>	<i>Impatiens necrotic spot virus</i>	INSV	Major	Indicator plants ( <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA See EPPO Diagnostic Protocol PM 7/34	<i>F. occidentalis</i> , <i>Frankliniella fusca</i>
<i>Potyvirus</i>	<i>Lettuce mosaic virus</i>	LMV	Major	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	Aphids
<i>Tospovirus</i>	<i>Tomato spotted wilt virus</i>	TSWV	Major	Indicator plants ( <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA See EPPO Diagnostic Protocol PM 7/34	<i>F. occidentalis</i> , <i>Thrips tabaci</i> , <i>T. setosus</i> , <i>F. fusca</i> , <i>F. intonsa</i> , <i>F. schultzei</i> , <i>Scirtothrips dorsalis</i>
<i>Dickeya</i>	<i>Dickeya (Erwinia) chrysanthemi</i>	–	Minor	Culturing and then confirmation by fatty acids analysis IF for screening	–
<i>Agrobacterium</i>	<i>Agrobacterium tumefaciens</i>	–	Minor	Culturing and then confirmation by PCR	–
<i>Verticillium</i>	<i>Verticillium dahliae</i>	–	Minor	Culturing, morphological identification	–

\*Pests categorized as major pests should be tested/checked/monitored during the entire propagation scheme, whereas it is recommended that the 'minor pests' are checked (if necessary with testing) at least during the selection and testing of the candidate nuclear stock plants.

#### Special requirements in the scheme

##### Useful life of Nuclear Stock Plant

1 year

##### Maximum number of generations of Propagation Stock

Propagation Stock I: 2

Propagation Stock II: 1

##### Pests especially requiring control

*Liriomyza* spp.



## Appendix 15

### Phlox

#### Species covered

*Phlox* spp. such as *P. drummondii*

#### Propagation

Cuttings, tissue culture, dividing, seed

#### Pathogens

Genus	Species	Acronym	Status*	Test method	Vector
<i>Alfavirus</i>	<i>Alfalfa mosaic virus</i>	AMV	Minor	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	Aphids
<i>Potexvirus</i>	<i>Alternanthera mosaic virus</i>	AltMV	Minor	Indicator plants (e.g. <i>Chenopodium quinoa</i> ), ELISA (can be detected with <i>Papaya Mosaic Virus</i> antiserum)	None, transmitted by sap
<i>Nepovirus</i>	<i>Arabid mosaic virus</i>	ArMV	Minor	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	<i>Xiphinema</i> spp.
<i>Luteovirus</i>	<i>Beet western yellows virus</i>	BWYV	Minor	ELISA	Aphids
<i>Cucumovirus</i>	<i>Cucumber mosaic virus</i>	CMV	Major	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	Aphids
<i>Tospovirus</i>	<i>Impatiens necrotic spot virus</i>	INSV	Major	Indicator plants ( <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA See EPPO Diagnostic Protocol PM 7/34	<i>F. occidentalis</i> , <i>Frankliniella fusca</i>
<i>Potyvirus</i>	Potyviruses	–	Major	Broad-spectrum ELISA, PCR†	Aphids
<i>Tobravirus</i>	<i>Tobacco rattle virus</i>	TRV	Minor	Indicator plants (e.g. <i>C. quinoa</i> ), PCR‡, ELISA§	<i>Paratrichodorus</i> and <i>Trichodorus</i> spp.
<i>Nepovirus</i>	<i>Tomato black ring virus</i>	TBRV	Minor	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	<i>Longidorus elongatus</i> and <i>L. attenuatus</i>
<i>Tobamovirus</i>	<i>Tomato mosaic virus</i>	ToMV	Minor	Indicator plants (e.g. <i>Nicotiana glutinosa</i> , <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA	None, spread by plant sap
<i>Tospovirus</i>	<i>Tomato spotted wilt virus</i>	TSWV	Major	Indicator plants ( <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA See EPPO Diagnostic Protocol PM 7/34	<i>F. occidentalis</i> , <i>Thrips tabaci</i> , <i>T. setosus</i> , <i>F. fusca</i> , <i>F. intonsa</i> , <i>F. schultzei</i> , <i>Scirtothrips dorsalis</i>
<i>Verticillium</i>	<i>Verticillium albo-atrum</i>	–	Minor	Culturing, morphological identification	–

\*Pests categorized as major pests should be tested/checked/monitored during the entire propagation scheme, whereas it is recommended that the ‘minor pests’ are checked (if necessary with testing) at least during the selection and testing of the candidate nuclear stock plants.

†e.g. Langeveld SA, Dore JM, Memelink J, Derks AFLM, Vlucht CIM van der, Asjes CJ & Bol JF (1991) Identification of potyviruses using the polymerase chain reaction with degenerate primers. *Journal of General Virology* **72**: 1531–1541.

‡Cornelissen BJC, Linthorst HJM, Brederode FT & Bol JF (1986) Analysis of the genome structure of tobacco rattle virus strain PSG. *Nucleic Acids Research* **14**(5), 2157–2169.

§Several sets of antisera should be used to detect various serotypes of the virus.

#### Special requirements in the scheme

*Useful life of Nuclear Stock Plant*

1 year

*Maximum number of generations of Propagation Stock*

Propagation Stock I: 2

Propagation Stock II: 1

*Pests especially requiring control*

*Ditylenchus dipsaci*

## Appendix 16

### Ranunculus

#### Species covered

*Ranunculus asiaticus* and its hybrids

#### Propagation

Tissue culture, dividing

Ranunculus are sexually (by seeds) and vegetatively (by cuttings and tissue culture) propagated.

The commercial propagation materials are:

- a) plants originated directly from seeds, or rooted cuttings originated from rhizomes, or plantlets originated by tissue culture
- b) rhizomes originated from plants of a), that are grown in propagator's premises.

This standard is intended to vegetatively propagated plants only.

#### Pathogens

Genus	Species	Acronym	Status*	Test method	Vector
<i>Cucumovirus</i>	<i>Cucumber mosaic virus</i>	CMV	Major	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	Aphids
<i>Tospovirus</i>	<i>Impatiens necrotic spot virus</i>	INSV	Major	Indicator plants ( <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA See EPPO Diagnostic Protocol PM 7/34	<i>F. occidentalis</i> , <i>Frankliniella fusca</i>
<i>Potyvirus</i>	<i>Ranunculus leaf distortion virus</i> † <i>Ranunculus mild mosaic virus</i> <i>Ranunculus severe mosaic virus</i>	Not approved yet	Major	Indicator plants ( <i>N. benthamiana</i> , <i>Ranunculus sardous</i> ), ELISA	Aphids
<i>Potyvirus</i>	<i>Ranunculus mottle virus</i>	RanMoV	Major	Indicator plants, ELISA	<i>Myzus persicae</i>
<i>Ophiovirus</i>	<i>Ranunculus white mottle virus</i>	RWMV	Major	ELISA	Not known
<i>Necrovirus</i>	<i>Tobacco necrosis virus</i>	TNV	Major	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	<i>Olpidium brassicae</i>
<i>Tospovirus</i>	<i>Tomato spotted wilt virus</i>	TSWV	Major	Indicator plants ( <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA See EPPO Diagnostic Protocol PM 7/34	<i>F. occidentalis</i> , <i>Thrips tabaci</i> , <i>T. setosus</i> , <i>F. fusca</i> , <i>F. intonsa</i> , <i>F. schultzei</i> , <i>Scirtothrips dorsalis</i>
<i>Fusarium</i>	<i>Fusarium oxysporum</i> f. sp. <i>ranunculi</i>	–	Minor	Culturing	–
<i>Monographella</i>	<i>Monographella cucumerina</i>	–	Minor	Culturing	–
<i>Pythium</i>	<i>Pythium sylvaticum</i>	–	Minor	Culturing	–

\*Pests categorized as major pests should be tested/checked/monitored during the entire propagation scheme, whereas it is recommended that the 'minor pests' are checked (if necessary with testing) at least during the selection and testing of the candidate nuclear stock plants.

†*Ranunculus leaf distortion virus*, *Ranunculus mild mosaic virus*, *Ranunculus severe mosaic virus* are new viruses and their name and acronyms are not approved yet by ICTV (International Committee for Taxonomy of Viruses).

#### Special requirements in the scheme

*Useful life of Nuclear Stock Plant*

1 year

*Maximum number of generations of Propagation Stock*

Propagation Stock I: 2 (for cuttings)

Propagation Stock II: 2 (for cuttings)

The explants can be maintained in vitro, to perform multiplication and rooting phases, for a maximum period of 18 months.

*Pests especially requiring control*

*Liriomyza* spp.

## Appendix 17

### Scaevola

#### Species covered

*Scaevola* spp. and hybrids

#### Propagation

Cuttings, tissue culture, seed

#### Pathogens

Genus	Species	Acronym	Status*	Test method	Vector
<i>Alfavirus</i>	<i>Alfalfa mosaic virus</i>	AMV	Minor	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	Aphids
<i>Cucumovirus</i>	<i>Cucumber mosaic virus</i>	CMV	Major	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	Aphids
<i>Tospovirus</i>	<i>Impatiens necrotic spot virus</i>	INSV	Major	Indicator plants ( <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA See EPPO Diagnostic Protocol PM 7/34	<i>F. occidentalis</i> , <i>Frankliniella fusca</i>
<i>Potyvirus</i>	Potyviruses	–	Minor	Broad-spectrum ELISA, PCR†	Aphids
<i>Tobamovirus</i>	<i>Tobacco mosaic virus</i>	TMV	Major	Indicator plants (e.g. <i>Nicotiana glutinosa</i> , <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA	None, spread by plant sap
<i>Tobamovirus</i>	<i>Tomato mosaic virus</i>	ToMV	Major	Indicator plants (e.g. <i>Nicotiana glutinosa</i> , <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA	None, spread by plant sap
<i>Tospovirus</i>	<i>Tomato spotted wilt virus</i>	TSWV	Major	Indicator plants ( <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA See EPPO Diagnostic Protocol PM 7/34	<i>F. occidentalis</i> , <i>Thrips tabaci</i> , <i>T. setosus</i> , <i>F. fusca</i> , <i>F. intonsa</i> , <i>F. schultzei</i> , <i>Scirtothrips dorsalis</i>
<i>Verticillium</i>	<i>Verticillium dahliae</i>	–	Minor	Culturing, morphological identification	–

\*Pests categorized as major pests should be tested/checked/monitored during the entire propagation scheme, whereas it is recommended that the 'minor pests' are checked (if necessary with testing) at least during the selection and testing of the candidate nuclear stock plants.

†e.g. Langeveld SA, Dore JM, Memelink J, Derks AFLM, Vlucht CIM van der, Asjes CJ & Bol JF (1991) Identification of potyviruses using the polymerase chain reaction with degenerate primers. *Journal of General Virology* **72**, 1531–1541.

#### Special requirements in the scheme

Useful life of Nuclear Stock Plant

1 year

Maximum number of generations of Propagation Stock

Propagation Stock I: 2

Propagation Stock II: 1

## Appendix 18

### Scrophulariaceae

#### Species covered

*Angelonia*, *Antirrhinum*, *Bacopa*, *Calceolaria*, *Diascia*, *Mecardonia*, *Mimulus*, *Nemesia*, *Penstemon*, *Sutera*, *Torenia*, *Verbascum*, *Veronica*

#### Propagation

Cuttings, tissue culture, seed depending on the species

#### Pathogen

##### Viruses

Genus	Species	Acronym	Status*	Test method	Vector
<i>Alfavirus</i>	<i>Alfalfa mosaic virus</i>	AMV	Minor	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	Aphids
<i>Carmovirus</i>	<i>Angelonia flower mottle virus</i> †	AFMoV	Minor (only for <i>Angelonia</i> spp.)	ELISA	None, spread by plant sap
<i>Luteovirus</i>	<i>Beet western yellows virus</i>	BWYV	Minor	ELISA	Aphids
<i>Fabavirus</i>	<i>Broad bean wilt virus 1 and 2</i>	BBWV-1, -2	Minor	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	Aphids
<i>Carlavirus</i>	<i>Chrysanthemum virus B</i>	CVB	Minor	ELISA	Aphids
<i>Cucumovirus</i>	<i>Cucumber mosaic virus</i>	CMV	Major	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	Aphids
<i>Tospovirus</i>	<i>Impatiens necrotic spot virus</i>	INSV	Major	Indicator plants ( <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA See EPPO Diagnostic Protocol PM 7/34	<i>F. occidentalis</i> , <i>Frankliniella fusca</i>
<i>Tymovirus</i>	<i>Nemesia ring necrosis virus</i> ‡	NeRNV	Major for <i>Diascia</i> Minor for all other species	Indicator plants ( <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA	None, spread by plant sap
<i>Potyvirus</i>	Potyviruses	–	Minor	Broad-spectrum ELISA, PCR§	Aphids
<i>Tobamovirus</i>	<i>Tobacco mosaic virus</i>	TMV	Minor	Indicator plants (e.g. <i>Nicotiana glutinosa</i> , <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA	None, spread by plant sap
<i>Tobamovirus</i>	<i>Tomato mosaic virus</i>	ToMV	Minor	Indicator plants (e.g. <i>Nicotiana glutinosa</i> , <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA	None, spread by plant sap
<i>Tospovirus</i>	<i>Tomato spotted wilt virus</i>	TSWV	Major	Indicator plants ( <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA See EPPO Diagnostic Protocol PM 7/34	<i>F. occidentalis</i> , <i>Thrips tabaci</i> , <i>T. setosus</i> , <i>F. fusca</i> , <i>F. intonsa</i> , <i>F. schultzei</i> , <i>Scirtothrips dorsalis</i>

\*Pests categorized as major pests should be tested/checked/monitored during the entire propagation scheme, whereas it is recommended that the ‘minor pests’ are checked (if necessary with testing) at least during the selection and testing of the candidate nuclear stock plants.

†This is a new virus, and its name is not yet agreed by ICTV (*Angelonia* flower break virus / *Angelonia* flower mottle virus).

‡This is a new virus, and its name is not yet agreed by ICTV.

§e.g. Langeveld SA, Dore JM, Memelink J, Derks AFLM, Vlugt CIM van der, Asjes CJ & Bol JF (1991) Identification of potyviruses using the polymerase chain reaction with degenerate primers. *Journal of General Virology* **72**, 1531–1541.

#### Special requirements in the scheme

##### Useful life of Nuclear Stock Plant

Usually 1 year, depending on the crop

##### Maximum number of generations of Propagation Stock

Propagation Stock I: 2

Propagation Stock II: 1

## Appendix 19

### Solanaceae

#### Species covered

*Calibrachoa*, *Brugmansia*, ornamental *Capsicum*. *Petunia* is covered by EPPO Standard PM 4/26 pathogen-tested material of *Petunia*

#### Propagation

Cuttings, tissue culture, seed depending on the species

#### Pathogens

Genus	Species	Acronym	Status*	Test method	Vector
<i>Alfavirus</i>	<i>Alfalfa mosaic virus</i>	AMV	Major	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	Aphids
<i>Fabavirus</i>	<i>Broad bean wilt virus 1 and 2</i>	BBWV1, 2	Minor	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	Aphids
<i>Carmovirus</i>	<i>Calibrachoa mottle virus</i>	CbMV	Major for <i>Calibrachoa</i> spp. Minor for other species	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	None, spread by plant sap
<i>Cucumovirus</i>	<i>Cucumber mosaic virus</i>	CMV	Major	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	Aphids
<i>Tospovirus</i>	<i>Impatiens necrotic spot virus</i>	INSV	Major	Indicator plants ( <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA See EPPO Diagnostic Protocol PM 7/34	<i>F. occidentalis</i> , <i>Frankliniella fusca</i>
<i>Potexvirus</i>	<i>Pepino mosaic virus</i>	PepMV	Minor	Indicator plants (e.g. <i>Datura stramonium</i> , <i>N. benthamiana</i> ), ELISA	None, spread by plant sap
<i>Tobamovirus</i>	<i>Pepper mild mottle virus</i>	PMMoV	Minor for <i>Capsicum</i> spp. only	Indicator plants (e.g. <i>Nicotiana glutinosa</i> , <i>C. quinoa</i> ), ELISA	None, spread by plant sap
<i>Tombusvirus</i>	<i>Petunia asteroid mosaic virus</i>	PetAMV	Minor	Indicator plants (e.g. <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA	None, spread by plant sap
<i>Pospiviroid</i>	<i>Potato spindle tuber viroid</i>	PSTVd	Minor	R-PAGE, DIG probe, PCR See EPPO Diagnostic Protocol PM 7/33	None, spread by plant sap
<i>Potexvirus</i>	<i>Potato virus X</i>	PVX	Minor	Indicator plants ( <i>Nicotiana</i> spp.), ELISA	None, spread by plant sap
<i>Potyvirus</i>	<i>Potato virus Y</i>	PVY	Major	Indicator plants ( <i>N. benthamiana</i> , <i>N. tabacum</i> ), ELISA	Aphids
<i>Potyvirus</i>	Potyviruses other than PVY	–	Minor	Broad-spectrum ELISA, PCR†	Aphids
<i>Tobamovirus</i>	<i>Ribgrass mosaic virus</i>	RMV	Minor	Indicator plants (e.g. <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA	None, spread by plant sap
<i>Nepovirus</i>	<i>Tomato black ring virus</i>	TBRV	Minor	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	<i>Longidorus elongatus</i> and <i>L. attenuatus</i>
<i>Tobamovirus</i>	<i>Tobacco mild green mosaic</i>	TMGMV	Major	Indicator plants (e.g. <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA	None, spread by plant sap
<i>Tobamovirus</i>	<i>Tobacco mosaic virus</i>	TMV	Major	Indicator plants (e.g. <i>Nicotiana glutinosa</i> , <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA	None, spread by plant sap
<i>Nepovirus</i>	<i>Tobacco ringspot virus</i>	TRSV	Minor	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA See EPPO Diagnostic Protocol PM 7/2	<i>Xiphinema americanum sensu lato</i>
<i>Cucumovirus</i>	<i>Tomato aspermy virus</i>	TAV	Minor	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	Aphids
<i>Tobamovirus</i>	<i>Tomato mosaic virus</i>	ToMV	Major	Indicator plants (e.g. <i>Nicotiana glutinosa</i> , <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA	None
<i>Nepovirus</i>	<i>Tomato ringspot virus</i>	ToRSV	Minor	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA See EPPO Diagnostic Protocol PM 7/49	<i>Xiphinema americanum sensu lato</i>
<i>Tospovirus</i>	<i>Tomato spotted wilt virus</i>	TSWV	Major	Indicator plants ( <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA See EPPO Diagnostic Protocol PM 7/34	<i>F. occidentalis</i> , <i>Thrips tabaci</i> , <i>T. setosus</i> , <i>F. fusca</i> , <i>F. intonsa</i> , <i>F. schultzei</i> , <i>Scirtothrips dorsalis</i>
<i>Ralstonia solana-cearum</i>			Minor	Diagnostic Protocol PM 7/21	

\*Pests categorized as major pests should be tested/checked/monitored during the entire propagation scheme, whereas it is recommended that the 'minor pests' are checked (if necessary with testing) at least during the selection and testing of the candidate nuclear stock plants.

†e.g. Langeveld SA, Dore JM, Memelink J, Derks AFLM, Vlucht CIM van der, Asjes CJ & Bol JF (1991) Identification of potyviruses using the polymerase chain reaction with degenerate primers. *Journal of General Virology* **72**, 1531–1541.

*Petunia vein clearing virus* (PVCV, *Caulimovirus*) is also a virus of phytosanitary concern for ornamental Solanaceae, but it is difficult to detect in routine inspections (test methods include indicator plant (*Petunia* 'Himmersröschen) and visual inspection).

### Special requirements in the scheme

*Useful life of Nuclear Stock Plant*  
1 year

*Maximum number of generations of Propagation Stock*  
Propagation Stock I: 2  
Propagation Stock II: 1

## Appendix 20

### Verbena

#### Species covered

*Verbena* spp. and hybrids

#### Propagation

Cuttings, tissue culture, seed

#### Pathogens

Genus	Species	Acronym	Status*	Test method	Vector
<i>Alfavirus</i>	<i>Alfalfa mosaic virus</i>	AMV	Major	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	Aphids
<i>Carmovirus</i>	<i>Angelonia flower mottle virus</i>	AFMoV	Minor	ELISA	None, spread by plant sap
<i>Fabavirus</i>	Broad bean wilt virus -1, -2	BBWV1, -2	Minor	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	Aphids
<i>Potyvirus</i>	<i>Clover yellow mosaic virus</i>	CIYMV	Minor	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	None, spread by plant sap
<i>Cucumovirus</i>	<i>Cucumber mosaic virus</i>	CMV	Major	Indicator plants (e.g. <i>C. quinoa</i> ), ELISA	Aphids
<i>Tospovirus</i>	<i>Impatiens necrotic spot virus</i>	INSV	Major	Indicator plants ( <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA See EPPO Diagnostic Protocol PM 7/34	<i>F. occidentalis</i> , <i>Frankliniella fusca</i>
<i>Tymovirus</i>	<i>Nemesia ring necrosis virus</i>	NeRNV	Minor	Indicator plants ( <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA	None, spread by plant sap
<i>Potyvirus</i>	Potviruses	–	Minor	Broad-spectrum ELISA, PCR†	Aphids
<i>Tobamovirus</i>	<i>Ribgrass mosaic virus</i>	RMV	Minor	Indicator plants (e.g. <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA	None, spread by plant sap
<i>Tobamovirus</i>	<i>Tobacco mosaic virus</i>	TMV	Minor	Indicator plants (e.g. <i>Nicotiana glutinosa</i> , <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA	None, spread by plant sap
<i>Tobamovirus</i>	<i>Tomato mosaic virus</i>	ToMV	Minor	Indicator plants (e.g. <i>Nicotiana glutinosa</i> , <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA	None, spread by plant sap
<i>Tospovirus</i>	<i>Tomato spotted wilt virus</i>	TSWV	Major	Indicator plants ( <i>N. benthamiana</i> or <i>N. occidentalis</i> ), ELISA See EPPO Diagnostic Protocol PM 7/34	<i>F. occidentalis</i> , <i>Thrips tabaci</i> , <i>T. setosus</i> , <i>F. fusca</i> , <i>F. intonsa</i> , <i>F. schultzei</i> , <i>Scirtothrips dorsalis</i>

\*Pests categorized as major pests should be tested/checked/monitored during the entire propagation scheme, whereas it is recommended that the 'minor pests' are checked (if necessary with testing) at least during the selection and testing of the candidate nuclear stock plants.

†e.g. Langeveld SA, Dore JM, Memelink J, Derks AFLM, Vlucht CIM van der, Asjes CJ & Bol JF (1991) Identification of potyviruses using the polymerase chain reaction with degenerate primers. *Journal of General Virology* **72**, 1531–1541.

#### Note 1:

In general *Verbena* is a difficult crop to test by ELISA, background noise may lead to false positives, low virus titres may lead to false negatives.

indicator plant research and it does not show symptoms in *Verbena*.

### Special requirements in the scheme

*Useful life of Nuclear Stock Plant*  
1 year

*Maximum number of generations of Propagation Stock*  
Propagation Stock I: 2  
Propagation Stock II: 1

#### Note 2:

*Verbena* latent virus (VeLV) is reported from *Verbena* but there is no test method commercially available.

*Angelonia* flower break (mottle) virus (AFMoV) was found recently, but its significance is yet to be determined. It may be considered at least as minor, since it is difficult to detect with