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SAGTEVRUGTEPLANTVERBETERINGSVERENIGING
DECIDUOUS FRUIT PLANT IMPROVEMENT ASSOCIATION

DPA F-1-3

TISSUE CULTURE FACILITIES

(1 August 2024)

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1. REGISTRATION OF TISSUE CULTURE FACILITIES

- 1.1 The facility must be registered with the Department of Agriculture (DoA) as a nursery if not only certified rootstocks are to be provided. Such a facility must meet the requirements prescribed in terms of the Plant Improvement Act, 2018 (Act No. 11 of 2018) before it can be registered with the DPA.
- 1.2 Plant improvement organisations that import tissue culture rootstock plant material or wish to propagate in a tissue culture facility for supply to nurseries must apply to register the relevant facility/s as foundation nursery/s (DPA D-1-1).
- 1.3 If a tissue culture facility wishes to directly import or propagate plant material, the relevant facility must apply to the Deciduous Fruit Plant Improvement Association (DPA) to register as a Plant Improvement Organisation (DPA C-1-1).
- 1.4 Facilities that apply must have the infrastructure and expertise as set out in Section 2.
- 1.5 Tissue culture facilities are registered with the DPA based on the equipment and the competence of the personnel doing the propagation at that stage. Any change or expansion in respect of the facility and expertise of staff must be reported to the DPA.
- 1.6 Registered facilities must have the necessary documented literature and references to follow the most effective protocol for each type of plant material.
- 1.7 The facility must consistently apply good laboratory practices and service records must be available as evidence that equipment is regularly checked and in good working order.
- 1.8 All registered facilities must be audited annually regarding the requirements and production of plant material.

2. REQUIREMENTS FOR FACILITIES

- 2.1 Tissue culture facilities that have a registered quarantine unit must ensure that the unit always meets the requirements of the DoA for such a unit.
- 2.2 The facility must also have a nucleus- / foundation tunnel where plants can be housed for propagation.
- 2.3 There must be clearly demarcated areas for the following activities:
 - 2.3.1 washing room;

- 2.3.2 storage area;
 - 2.3.3 media preparation room;
 - 2.3.4 media storage;
 - 2.3.5 inoculation;
 - 2.3.6 growth room;
 - 2.3.7 plant transfer area;
 - 2.3.8 primary hardening area (acclimatisation); and
 - 2.3.9 secondary hardening area.
- 2.4 There must be limited access to all areas where plant material and media are handled and stored.
- 2.5 The facility must have sufficient emergency power to ensure that power supply is not interrupted.
- 2.6 The growth medium used when transplanting plantlets from the laboratory must be sterile.
- 2.7 All water sources for both the laboratory as well as the hardening units and greenhouses must be free of pests and vectors of deciduous fruit diseases.

3. REQUIREMENTS FOR PLANT MATERIAL

3.1 Imported plant material

- 3.1.1 Imported tissue culture material is inspected by DoA and then released to the approved tissue culture facility, to be housed in the quarantine unit.
- 3.1.2 If the material comes from an accredited facility and meets all the phytosanitary requirements, the material can be released from quarantine quickly.
- 3.1.3 In cases where the material does not come from an accredited facility, hardened plants must be provided to the quarantine station to carry out the necessary tests and investigations before the shipment can be released.
- 3.1.4 The plant material referred to in 3.1.3 will not qualify for certification before all the necessary tests and evaluation have been completed locally.
- 3.1.5 In cases where plant material (mass imports) has been imported from an accredited facility (3.1.2) and where proof can be provided that the material in question also meets the phytosanitary requirements of the Scheme, an application can be made to include the consignment in question in the Scheme.

3.1.6 The plant material must meet the following requirements:

- a) The variety must be included in the Official Variety List;
- b) the clone must be registered;
- c) the origin must be a certified source plant at an accredited overseas facility.

3.1.7 If plant material comes from a cultivar that is not yet on the Official Variety List, an application must have already been made for placement on the list and proof must be provided that dispensation for further propagation has been granted.

3.1.8 In the case of mass imports, a minimum of 0.1% (at least 10 plants) must be tested during the hardening process for the absence of the scheme viruses as well as for varietal authenticity

3.2 Local plant material

3.2.1 The PIO must supply certified material from a nucleus tunnel to the tissue culture facility.

3.2.2 In cases where a nucleus plant is not available, a source plant must first be established in the nucleus- /foundation unit of the facility.

3.2.3 The source plants in 3.2.1 and 3.2.2 must be treated with the necessary cleaning agents to ensure that the risk of contamination of the *in vitro* cultures is minimised.

3.2.4 Source plants must be replaced at least every five years.

3.2.5 The facility takes plant material from these source plants and establishes, after a process of surface sterilisation, "mother ex-plants" that will serve as a source for the subsequent initiations.

3.2.6 All source plants must be PCR tested for all scheme viruses.

3.2.7 The varietal integrity of cultivars must be confirmed by DNA before propagation.

3.2.8 Where successive mass propagation is done, a minimum of 0.1% (at least 10 plantlets) must be tested during the hardening process for the absence of the scheme viruses as well as for varietal authenticity.

4. INCLUSION IN THE SCHEME

- 4.1 Thorough records must be kept of each step of the propagation process in terms of clones and numbers within the laboratory as well as of all plantlets that are transferred to the mist bed for primary hardening.
- 4.2 These records will be audited annually by Plant SA.
- 4.3 Once plant material is transferred from the primary hardening phase to the secondary phase, the material is officially included in the Scheme and the facility must provide the DPA with a completed nursery return (DPA D-3-4).
- 4.4 The requirements as set out in the Standard Operating Procedure for Nurseries (DPA D-1-1) must be followed from this stage forward for the plant material.