

## **BLACK PEPPER- TISSUE CULTURE – (BPTC)- STANDARDS**

### **I. Applications and Amplification of General seed Standards for BPTC**

- a. The General Seed Certification Standards are basic and, together with the following specific standards constitute the standards for approval of BTC. As the name implies, these standards are applicable to tissue culture multiplied under laboratory and greenhouse conditions as laid here.
- b. The General Standards are amplified as follows to apply specifically to the BPTC.

#### ***1. Eligibility requirements for BPTC production:***

- i. All micropropagation and greenhouse facilities must be approved as per standards/ guidelines set by the competent authority. These must have a changing area between double doors.
- ii. Laboratory and greenhouse facilities used for production of plantlets shall be maintained free of pests or vectors of banana pathogens. Failure to keep such pests under control may cause rejection of all lots maintained in the facility. All potting or growth media shall be sterile. Water sources used in the laboratory or greenhouse operation shall be treated or otherwise rendered free of all possible pathogens by the applicant.
- iii. Hygienic conditions shall be strictly observed during micropropagation, potting, planting, irrigating, movement and use of equipment and other laboratory and greenhouse practices to guard against the spread of diseases or pests in the facilities used for banana plant multiplication.
- iv. The greenhouse (protected environment) must be “insect proof” and be equipped with a double-door entrance, provision for footwear disinfection prior to entering the protected environment and insect proof ventilation screening on intakes and exhaust openings. The persons entering the protected environment should use Wellington boots (plastic boots) and change lab-coat in the changing area to reduce the chances of inadvertent introduction of vector insects clinging to clothes

- v. The material being initiated must be of a notified variety and confirmed identity. It must be duly documented with respect to origin.
- vi. All samples of black pepper varieties being initiated should be tested in an accredited laboratory and should be free of viruses (CMV, badnavirus phytoplasma) and other endophytic or epiphytic bacteria and fungi
- vii. The basic material for sub-multiplication need to be obtained afresh from the nodal organization as soon as the maximum permitted number of passages (as confirmed by DNA fingerprinting) of shoot multiplication with old cultures has been completed.
- viii. On application for inspection, the mother cultures as developed above are eligible for certification. The micropropagation facility to be inspected must have been approved by the competent Authority. All stocks must have a valid variety identification and disease testing report at any time during multiplication process.

*In vitro multiplication of an imported variety or a non-notified variety can be taken up by the industry exclusively for export purposes. Such varieties, however, should be introduced following the approved guidelines of Government of India.*

## **2. Source of Seed:**

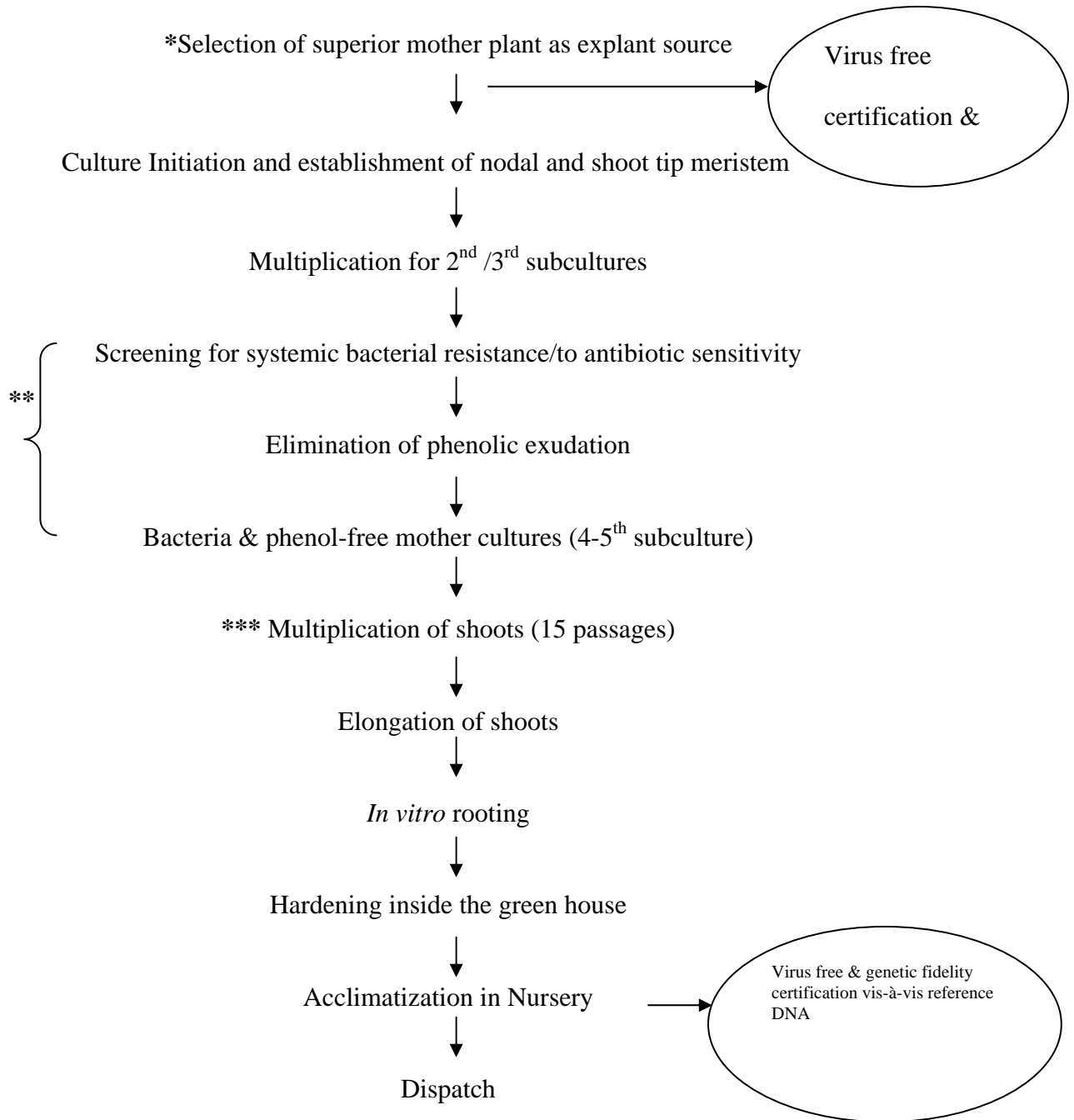
- i. The facility should use recognized aseptic initiation and propagation procedures (i.e. follow procedures and use equipment, which will maintain sterile conditions as per standard tissue culture norms).
- ii. The initiating facility must maintain following information on each variety for review and audit by the competent authority at least once in a year: variety identification, date of initiation, origin and testing results from accredited laboratory.
- iii. Tests must be carried out on a minimum of 0.1% (minimum ten) plantlets for each variety by an accredited laboratory. Such tests will be valid as long as cultures of that particular batch are under production. No plant should contain (CMV and Badnavirus) and other endophytic or epiphytic bacteria and fungi.
- iv. Valid pathogen testing results are required at the 2<sup>nd</sup>/3<sup>rd</sup> subculture stage prior to the bulking up of the cultures.
- v. The guidelines for production of tissue culture plant is at Appendix-I

### **Minimum Quality Standards for growing of plants inside greenhouses/polyhouses**

The following requirements must be met for production of plantlets :

- i. Effective sanitation practices including insect and disease monitoring and prevention must be adhered to.
- ii. No field-produced black pepper plants can be grown in the protected environment (greenhouse/polyhouse) along with tissue cultured plants.
- iii. Varieties must be separated by physical barriers (such as proper tagging), which will prevent varietal mixture.
- iv. Before dispatch to the farmers, the tissue-cultured plants growing in the nursery should be tested for the absence of the viruses (CMV, Badnavirus) and clonal uniformity. For establishing clonal fidelity, the sample size should be 0.1% of the batch size with a minimum of 10 plants.
- v. If testing performed by an accredited laboratory reveals the presence of banned viruses, fungus or bacteria the tissue-cultured plants should not be dispatched from the premises of the production lab and the entire material should be destroyed.
- vi. The concerned laboratory/agency producing the tissue culture raised material should issue a certificate to the effect that BPTC have been produced as per guidelines
- vii. The agency producing BPTC will follow the labelling procedures as given at Appendix-II

**Procedure and standard parameters for production of Black pepper by tissue culture**



\* *Plants should be of superior quality in terms of growth, disease / pest resistance, drought tolerance, constant bearing (confirmed for at least three consecutive years), high yield (fresh and dry weight), oil and oleoresin content etc. The explant should be healthy and free from microbial infections. One set of mother plants must be maintained in the glass house as reference sample.*

\*\* *Since black pepper tissue culture is frequently confronted with endogenous bacterial contamination and phenolic exudation, these should be eliminated using appropriate method.*

\*\*\* *In black pepper the number of passages can be up to 15 for subculture of shoots.*

**Labelling Black Pepper-Tissue Culture (BPTC)**

1. BPTC shall be supplied in containers. A cloth-lined label of 12cm x 6 cm cm containing following information shall be affixed on the container

**Crop** : **Black pepper**                      **Lable No.:**

**Variety**

**Class of material** : **BPTC**

**Lot No.** : **Batch**

**Accredited test laboratory  
and certification reference note** :

**Date of certification** :

**Production Agency** :  
**(Name and address)**

**‘The container should also have printed on it the kind, variety and name of Institution’**

2. The label shall be rubber stamped with signature, name and designation of the concerned Agency. Colour of the label shall be diagonally yellow No. 356 (IS 5-1978) and opaline green (IS No. 275)
3. BPTC producing Agency shall maintain the account of labels printed and issued