

## **BAMBOO- TISSUE CULTURE – (BaTC)- STANDARDS**

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

- a. The General Seed Certification Standards are basic and, together with the following specific standards constitute the standards for approval of BaTC. 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 BaTC.

#### ***1. Eligibility requirements for BaTC 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 bamboo 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 bamboo 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 bamboo varieties being initiated should be tested in an accredited laboratory and be free of 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% (at least ten) plantlets for each variety by an accredited laboratory. Such tests will be valid so long as cultures of that particular batch are under production (subject to a maximum of 15 passages). No plant should contain endophytic or epiphytic bacteria and fungi. Generally, viral infection is not seen in bamboos. However, there are reports that suggest infection by Bamboo Mosaic Virus (BaMV) at the nursery stage in India.

- 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.

### **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 bamboo 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 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 BaTC have been produced as per guidelines
- vii. The agency producing BaTC will follow the labelling procedures as given at Appendix-I

### **Nursery Development**

The plants to be used for field trials would be supplied either in polythene bags or bare-rooted (to save the transportation cost). In both cases the plants should be kept in the nursery till they have recovered from the transportation shock. While the plants in polythene bags may be kept directly in the nursery, the bare-rooted plants should be transferred to the polybags prior to their transfer to the nursery. The plants should be kept in the nursery for a minimum period of **10 days**. However, many a times the plants supplied are not of planting height. In such a situation it would be necessary to prolong the stay of plants in the nursery (till the

plants are at least **18 inches** in height) before their transfer to the planting site in an open field.

**Requirements of a bamboo nursery (holding area)**

- The nursery site should be on level ground and well drained
- It should be as close as possible to the plantation site
- It should have all necessary facilities for irrigation of plants
- The site should be protected from animals

**Managing a bamboo nursery**

- In the nursery the plants of different species should be kept in separate beds to avoid any mixing. If there are more than one genotype for each species, then it would be desirable to keep the plants genotype-wise.
- The approximate size of the nursery bed could be (8-10 m x 1-1.5 m)
- As much as possible, the beds should be prepared where there is some protection (shade of a tree/thatch) for the plants from direct sunlight
- The beds should be leveled so that there is no accumulation of water
- Each bed should be properly labeled so that there is no mixing of plants
- In the nursery the plants should be irrigated periodically, and care should be taken that they remain free of diseases
- Only healthy plants of uniform size (approximately) should be used for field trials, particularly experimental trials

**Attributes of primary and secondary hardened tissue cultured bamboo plants**

<b>S. No.</b>	<b>Trait</b>	<b>Primary hardened tissue cultured plant</b>	<b>Secondary hardened tissue cultured plant</b>
1.	Minimum height of the shoot	5 inches	18 inches
2.	Minimum number of shoots	> 3	> 4
3.	Minimum number of leaves per shoot	> 4	> 8

4.	Root system	Well developed	Well developed
5.	Rhizomes	Small rhizomes in the initial stages of development	Proper rhizomes
6.	Minimum age of the plant	1 month	> 3 months

## Labelling Bamboo-Tissue Culture (BaTC)

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

### **Certified Tissue Culture Raised Quality Plants/Propagules**

**NCS-TCP** Certified Tissue Culture Raised Quality Plants/Propagules

Certificate of Quality No.: .....

Label No. : .....

Name of Production Facility: .....

Botanical Name: .....

(Common Name): .....

Certification No. and validity: .....

Variety: .....

of Certificate of Recognition: .....

Batch No. & Batch Size: .....

Contact person and Designation: .....

Stage of Tissue Culture Plants:

Address with phone number: .....

In agar  Ex-agar  Hardened

Date of Issue: .....

Bar Coding :

Name/Sign/Stamp of ATL with date: .....

**‘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. BaTC producing Agency shall maintain the account of labels printed and issued