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Quality control of bamboo planting material through state-of-the-art technology

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Abstract

Recent developments in utilization of bamboo biomass for an increasing array of engineered bamboo products and as feedstock for bioenergy has greatly increased the potential demand for raw material in the coming years. This can only be met by expanding the extent of plantations around the world and increasing the productivity through interventions like use of quality planting material derived from genetically superior clumps and management of plantations on scientific principles. With a large diversity of species and relatively low level of genetic improvement due to the recent history of domestication and constraints in conventional breeding, the quality of bamboo planting material being used in plantations of bamboo is always called into question. Modern technology has the potential to provide solutions for many of the constraints that come in the way of efficient propagation and genetic improvement. DNA barcoding offers the solution for the vexing problem of identification of species especially at the juvenile stage. DNA fingerprinting can ensure that clonal fidelity and identity is maintained during large-scale propagation. Micropropagation is already being used commercially for bamboo propagation but is limited to a few species due to constraints such as incidence of microbial contamination, recalcitrance of tissues to in vitro culture establishment and rooting. This paper examines the potential of current technologies to support a certification scheme for quality control of planting stock as a means of ensuring that productivity of future bamboo plantations is enhanced. The array of modern techniques that can form the basis of such a scheme for accreditation of bamboo nurseries that national authorities can adopt, as has been recently done in India, to ensure that planted bamboo forests of the future are established on sound scientific footing and productivity of bamboo is enhanced several fold to meet the projected demand by growing industry.

Keywords Certification Scheme; Planting Material; DNA Barcoding; DNA Fingerprinting; Micropropagation

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1. Introduction

Recent advancements in technology has resulted in an array of industrial applications and products utilizing bamboo. These have a great potential in the coming years due to the fast renewable nature of bamboo biomass. Bamboo exhibits a great diversity across the tropical clumping and temperate running species that can be grown in almost all the continents of the world save the Antarctica. The increased demand for bamboo ligno-cellulosic biomass and fiber expected in the immediate future can only be met if the extent of bamboo plantations are significantly increased around the world. There are indications that this is indeed happening with bamboo plantations making inroads into Africa and large-scale planting of bamboo being envisaged in countries of Asia and South and Central America.

The productivity of bamboo plantations is an issue that will be sooner or later be brought to the fore when the initial emphasis on increase in acreage is over. Use of quality planting material will be examined and the constraints that prevent conventional breeding approaches to be applied to bamboo will receive attention. The long-life cycles of 30 to 120 years of bamboo species and the lack of simultaneous flowering of bamboo species are the hurdles to undertaking breeding as in agricultural crops or even forest trees. Bamboos have a fairly recent history of domestication and therefore selection has not progressed to the same extent as in many other commercial species.

It will therefore require the application of an array of scientific techniques to overcome the hurdles to genetic improvement in this group of plants that is poised to become the new industrial crop of the future. Modern biotechnological tools that have appeared on the scientific horizon in recent decades hold much promise in this regard. A recent attempt to integrate these technologies into a national certification scheme (National Bamboo Mission 2019) is an example of what can be achieved through use of known and emerging technologies to the bamboo nurseries engaged in producing planting stock for the bamboo plantations.

2. Technologies to the rescue

2.1. DNA Barcoding

With the advent of molecular techniques in biology it has now become possible for selected techniques to be used for identification at species level. DNA barcoding in particular is taking the field of taxonomy by storm and is fast becoming the norm for precise identification and new species discovery. The concept of using short stretch of DNA sequences as barcode for species identification had been proposed using the CO1 barcode region in animals (Hebert et al. 2003) but it soon became apparent that this region is ineffective for the purpose in plants.

Subsequently, based on the discrimination power across various angiosperm genera, a core barcode comprising matK+rbcL together with psbA-trnH as well as the nuclear ribosomal internal transcribed spacer region (ITS2) was recommended for barcoding in plants (CBOL 2009; Hollingsworth et al. 2009). It is however becoming increasingly clear that the recommended regions do not work efficiency in several groups of plants and the search is to be continued for more suitable barcodes either singly or in combinations. In bamboo the importance of barcoding has been emphasised by Sijimol et al (2014) and following a thorough study, Dev et al (2020) have come up with a barcode region that discriminates the major commercially important bamboo species in India. To develop a universal barcode for all bamboo still remains a challenge that requires research and the recent trend of using whole genome sequencing has given some hope that plastid genome will yield some results.

Use of DNA barcoding can therefore currently supplement the conventional morphology-based identification which is the domain of expert taxonomists and an area constrained by the lack of flowers in bamboo for most of the life cycle and culm sheaths for most of the year. The guidelines released by the National Bamboo Mission (2019) anticipated the provision of use of DNA barcoding in the precise identification of bamboo species before the selection of mother clump used for propagation and production of planting stock in nurseries.

2.2. DNA Fingerprinting

Clonal fidelity of planting stock was not an important issue when seedlings or vegetatively propagated plants were the main means of propagation. When tissue culture techniques achieved prominence and micropropagation became the accepted means of large-scale production of plants, the risk of somaclonal variation in some of the techniques was considered serious enough to introduce the genetic fidelity test using molecular markers. The DNA finger prints of the mother plants and the samples taken per batch of micropropagated plantlets should match for the batch to be accepted for certification. In India it is the Department of Biotechnology that administers the certification scheme (DBT 2016). A section on bamboo tissue culture is included in the certification system although the industry predominantly uses the axillary bud proliferation pathway that is relatively free of genetic variation. DNA fingerprinting is also a technique that ensures that clonal identity is known. Thus, inadvertent mixing of batches or misuse of clones in the nursery business can be avoided.

2.3. Micropropagation

The use of tissue culture techniques for propagation of bamboo is more than half a century old and has come of age in recent years with several commercial companies using it on a routine

basis around the world. The advantages of high rates of multiplication, uniformity of plantlets produced, disease and pest free status of plantlets are in contrast to the slow and cumbersome vegetative propagation procedures that result in bulky plants that are difficult to transport. While a large number of papers in literature (Sandhu et al. 2017) described micropropagation protocols of the important commercial species, only a limited number of them have been scaled up. This points to the hurdles faced in tissue culture that are not addressed in the scientific literature adequately. The high incidence of endophytic bacterial and fungal contamination in bamboo in the early stages of establishment is a typical example. There have been several approaches that are used to address this issue including the approach of biostasis that keep the microbial flora suppressed without total elimination. The induction of microrhizomes in micropropagated bamboo plantlets is another approach that has not received due attention in the industry. Presence of rhizomes in the plantlets gives an extra boost to the vigour and increases the chances of survival and establishment in the field on transfer. Control over in vitro flowering which results in senescence of shoot cultures has been achieved by avoiding stress factors. Other factors that control the recalcitrance of some of the species to in vitro culture have been overcome by use of newer plant growth regulators and modifications in culture procedures.

3. Discussion

A national level certification scheme to regulate the nursery industry will be needed to be implemented since the state-of-the-art of conventional methods for species and clonal identification and propagation is beset with problems that challenges the concept of quality. Interventions with the use of some of the latest techniques such as DNA fingerprinting and barcoding will help in precise identification both as a quality control tool as well as to implement IPR in the commercial context. While the use of molecular markers has found application in selection of plants with desirable agronomic traits (marker assisted selection) in several agricultural and horticultural crops, their use in bamboo genetic improvement is yet to take off. The basic principles remaining the same and bamboo being a grass, it is anticipated that soon we shall see an increased use of such techniques in selection of genetically superior bamboo. Recent advances in whole genome sequencing has opened up a vast amount of sequence data that given the homology with crops of the grass family like rice, wheat and corn, will accelerate the process of selection of superior clumps that can be mass multiplied for production of quality planting stock. Quality control through use of molecular techniques, of the tissue culture procedures used for large scale propagation also ensures that genetically true to type plants are produced.

When integrated with the activities in bamboo nurseries where the farmers and planters obtain the planting material, these modern techniques will ensure that only the certified mother clumps are multiplied. Standards that define the minimum requirements of a quality bamboo plant have been outlined in the Guidelines issued by National Bamboo Mission as well as the Department of Biotechnology in India. Accreditation of bamboo nurseries and bringing it under the purview of a central monitoring agency will help enforce the latest in technology and facilities to ensure that the quality of plant produced and sold meets rigorous standards so that the plantations are established with the best of planting material and buyers have the assurance that plants are of the correct species and clonal and known genetic antecedents.

Conflict of Interest

The authors declare there is no conflict of interest

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