THE APPLICATION OF BIOTECHNOLOGY IN BIODIVERSITY CONSERVATION

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ABSTRACT

The continuous loss of biodiversity has posed a serious threat to the survival of mankind. Worldwide one third of the plant species are threatened due to several reasons. As the conservation of biodiversity is a global concern, several strategies have been adopted in understanding and conserving plant diversity throughout the world. Both *ex situ* and *in situ* methods of biodiversity conservation are equally important. Though it is generally believed that biotechnology has adverse effects on biodiversity, but in fact, biotechnology offers new means of improving biodiversity. It is now well recognized that an appropriate conservation strategy for a particular genotype requires combining approach of *ex situ* and *in situ* techniques according to the need of the program. As it is known fact that genetic variability is the main perquisite of the survival of any plant species in their natural habitat, so study of genetic diversity in conserved germplasm is important and application of different biotechnological process playing a promising role. *In vitro* plant propagation is a helpful technique in the conservation of genetic diversity of all types of plants (including rare, threatened and endangered plants) in a rapid and reliable way by maintaining the same clone or stock of plant material. The review presents the use of biotechnology in improving *ex situ* conservation process to maintain biodiversity.

Keywords: Biodiversity, Biotechnology, Conservation, endangered.

INTRODUCTION

Biodiversity is the mass of different living beings in a particular ecosystem or on the whole earth. It exists in three different levels; genes, species, and ecosystems. Each of the components has its own composition, structure and function (Redford & Richter, 2001; Noss, 2005). Biodiversity can also be defined as the variety of all forms of life and it includes the abundance and richness of species. Biodiversity is the variability among living organisms: within and between species and ecosystems. However, there is no overarching indicator for biodiversity (Gregory, 2005).The most important determinant of biodiversity is habitat, which is the land cover. Biodiversity is considered as the foundation of agriculture being the source of all crops and livestock species that have been domesticated and bred since the beginning of agriculture approximately 10,000 years ago (Cooper, 2005). Changing the land cover in an undesired direction for biodiversity is considered as a pressure. Biodiversity loss is occurring in many parts of the globe, often at a rapid pace. This can be measured by loss of individual species, group of species or decreases in number of individual organisms. One of the tools that can be used to enhance biodiversity is biotechnology (Nill, 2002). Biotechnology is as the art of utilizing living organisms and their products for the production of food, drink, medicine or for other benefits to the human being, or other animal species. Biotechnology existed long before there was a special word for it. Biotechnology in one form or another has flourished since prehistoric times. When the first human beings realized that they could plant their own crops and breed their own animals, they learned to use biotechnology. More recently, cross-pollination of plants and cross-breeding of animals were macro-biological techniques in biotechnology, used to enhance product quality and/or meet specific requirements or standards. Even though, the discovery of microorganisms, antibiotics, causes of infectious diseases, and immunizations could probably be reckoned among significant discoveries; the most modern techniques in biotechnology get their existence to the discovery of DNA and several techniques necessary for gene cloning. This paper presents a review on the conservation of biodiversity, the important roles of biotechnology for the conservation of biodiversity and some potential ricks of modern biotechnology to biodiversity.

Biodiversity Loss

It has been estimated that one third of the global plant species are threatened in different level according to International Union of Conservation of Nature (IUCN, 2013). The increasing awareness and great concern about global biodiversity conservation, the United Nations (UN) declared the current decade (2011-2020) as the "Decade of Biodiversity' and has set 2020 as the target for restoring at least 15% of degraded ecosystems as well as conserving 17 and 10% of terrestrial and inland water and marine and coastal areas respectively (Tscharntke et al., 2012; SCBD, 2014). Global concern about the loss of valuable genetic resources stimulated many new programs for the conservation, protection and management of natural resources and wildlife. Losses of biodiversity are undoubtedly occurring in many parts of the globe, often at a rapid pace. These losses require countermeasures such as an increased effort towards conservation by many different means. In a given location, the loss will often reflect the degradation or destruction of whole ecosystem. The loss of biodiversity can be measured by a loss of individual species, groups of species or decreases in numbers of individual organisms.

CBD ranked the priority of threats to global biodiversity in the following manner:

- Habitat loss: probably the most serious of all threats to biodiversity (most of it through the expansion of cultivated land). Habitat loss comes not only from taking more land under the plough, but also from expanding cities and road building. Human activities such as pollution, human population pressure, agricultural practices, life style change etc. (Opdam & Wascher, 2004). The United Nations Food and Agricultural Organization (FAO) assuming that global population is approaching towards 9.1 billion in 2050 and there is a need of 70% increase in food production (Godfray et al., 2010). In this current situation, undertaking of effective as well as productive agricultural land uses has raised a global challenge of conserving biodiversity (Tscharntke et al., 2012).

- Introduction of exotic species: The second most important reason for loss of biodiversity is invasion by exotic plants and animals. Knowingly, or unknowingly, imported plant species threaten the native ones by being highly competitive and often by lacking local predators, such as insects or birds. It is estimated by **Sukopp & Sukopp (1993)** that one in ten imported plants may spread in a modest way and that one in a hundred may turn into a nuisance weed. Exotic biological control agents are often introduced into agricultural ecosystems on purpose, in order to control pests or weeds without resorting to chemical controls agents. Whilst there are some success stories, Strong pointed out that such systems may also go wrong. One example is the introduction of the seven-spot ladybird which was intended to fight the Russian wheat aphid. The consequence, however, was the disappearance of the native ladybirds, for which the seven-spot import was a competitor and an actual predator. According to the World Conservation Union (IUCN), habitat destruction followed by Invasive Alien Species (IAS), has been a major cause of extinction of native species. This invasion by biological control agents has caused a significant loss of biodiversity. Globally, the damage caused by invasive species has been estimated to be £1 trillion per year – close to 5% of global GDP (Lowe *et al*., 2000).
- Flooding, drought, climate changes, salinization and so on, all of which can be either natural or man-made (Ammann, 2005).

ROLE OF BIOTECH IN BIODIVERSITY CONSERVATION

Biodiversity conservation is the protection, restoration and sustainable management of wildlife and natural resources such as forests, water and the biological diversity within it. Through the conservation of biodiversity not only the survival of many threatened species and habitats can be ensured, but also these valuable resources will be secured for future generations and the well-being of eco-system functions protected. Biotechnology is a set of techniques by which human beings modify living things or use them as tools. In its modern form, biotechnology uses molecular biology techniques to understand and manipulate the basic building blocks of living things. The earliest biotechnology, however, was the selective breeding of plants and animals to improve their food value. Modern biotechnology offers new means of improving rather than threatening biodiversity. If properly tested for both risks and benefits to humans and the environment, transgenic crops are more likely to increase agricultural biodiversity and help maintain native biodiversity rather than to endanger it, in contrast to the claims of many environmental groups. Such applications need to be judged by the criteria of improved sustainability and compared to current as well as alternative farming practices (Ammann, 2005).

Biotechnology is presently used for the conservation, evaluation, and utilization of biodiversity particularly for important crops (Singh, 2000).

Biotechnology plays an immense role in biodiversity conservation such as listed below;

1. **Plant Tissue culture**

Plant tissue culture (PTC) is a very effective method of cloning of plant material and to develop disease-free clean plant stock. It is a quick, season independent and efficient *in vitro* technique to propagate plants under sterile micro environment. Evert plant cell has the power of cellular totipotency to be differentiated into whole plant in the process of plant tissue culture methodology. Different techniques in PTC may offer certain advantages over traditional methods of propagation for assembly, proliferation, preservation and storage of plant genetic resources (Bunn et al., 2007). It has great importance in the crop improvement program which is facing the increasing depletion of natural resources. Moreover, tissue culture techniques can be applied in germplasm conservation of important plants species of economic and medicinal values. It can be applied for regenerating different clean disease-free stock of plants in the field of agriculture, horticulture, floriculture and pharmaceutical industry (Fischer et al., 2004). Tissue culture is a useful technique to preserve somatic embryos which can be applied in the medium and long-term conservation process. PTC is also a great source of creating variations through the development, selection and isolation somaclones commonly known as somaclonal variations. Thus, biotechnology leads to the production of a new category of germplasms, clones of special category, elite cell lines and genetically transformed material with desired traits. The cultivation and conservation of the new germplasms in the changed environmental situation can be able to add some specific impact in changed environmental situations. Rapid and mass propagation of plant species and their long-term germplasm storage can be achieved in a small space within short time period, with no damage to the existing population using PTC techniques. Plant material can be produced throughout the year without any seasonal limitation. Large numbers of uniform and disease-free, virus-free plants can be produced from very small portions of the mother plant due to the aseptic nature of tissue culture technique (Sharma & Sharma, 2013). Genetic resources of recalcitrant seeds which are difficult to germinate, vegetatively propagated plants, rare, threatened plant species, elite crop varieties and some genetically modified plant materials can be efficiently multiplied and stored in long term basis by using *in vitro* techniques (Lidder & Sonnino, 2012).

Tissue culture has been applied to diverse research techniques such as viral elimination, clonal propagation, gene conservation, *in vitro* fertilization, mutation, induction for genetic diversity, genetic transformation, protoplast isolation and somatic hybridization, secondary metabolite production and other related techniques (Pathak and Abido, 2014).

2. Micropropagation and Cloning

In vitro clonal propagation method commonly known as “micropropagation” helps in the mass production of plant propagules from any plant part or cell. Micropropagated propagules are used to raise and multiply the stock plant material in micro environment. Micropropagation and cloning of plant tissue based on different explants is commonly used to conserve different endangered plants (Castellanos et al., 2008; Sadeq et al., 2014). Micropropagation technique assists in the rapid, season independent, continuous propagation, maintenance and storage of rare and endangered plants by using any plant parts as explant source (Sarasan et al., 2006; Chandra et al., 2010).

3. Somatic Embryogenesis and Organogenesis

The development of somatic embryo by the differentiation of a single somatic cell or tissue to regenerate large number of plants at the same time is very commonly used technique of somatic embryogenesis in plant tissue culture. Somatic embryogenesis and organ development through organogenesis from various culture of explants are the most commonly used technique applied to regenerate several endangered plants for the purpose of conservation (Sadeq et al., 2014).

4. Growth Restriction

One of the uses of the practical of a plant tissue culture method in germplasm conservation is to reduce the frequency of subculture during the process of culture. Development and use of several slow growth culture situations in modified culture media help to reduce the subculture frequency (Bunn et al., 2007). The addition and alteration of growth regulators, modification of salt concentration of culture media, high or low concentration of sugars in the growth media, addition of osmotically active compounds and sometime modulation of some external factor such as low temperature are used in slow growth strategy (Reed & Chang, 1997). Tropical species are mostly cold-sensitive and have to be stored at high temperature. *In vitro* cultivation of *Musa* sps. can be stored up to 15 months at 15ºC (Banerjee & de Langhe, 1985). While other tropical species such as cassava are more cold-sensitive, so their shoots can be stored at temperatures higher than 20ºC (Roca et al., 1984). *In vitro* slow growth storage technique is routinely used for medium-term conservation of numerous species both from tropical and temperate origin and endangered species (Pence et al., 2002).

5. Cryopreservation

Cryopreservation is one of the biotechnological method of *ex situ* plant conservation and applicable for long term storage of plant genetic material. Cryopreservation is extremely helpful method to conserve rare, endangered, threatened plant species (Dussert et al., 1997; Zhao et al., 2008; Paunescu, 2009). Moreover, for long-term storage of cultured plant materials require the use of ultra-cold storage methods. Most of the experimental systems used in cryopreservation (cell suspensions, calli, shoot tips, embryos etc.) contain high amount of cellular water and are thus very sensitive to freezing injury. So, artificial dehydration procedure is important step although classical technique. These involve freeze induced dehydration, while new techniques are based on vitrification (Zhao et al., 2008). The principle is bringing the plant cell and tissue cultures to a non-dividing or zero metabolism stage by subjecting them to supraoptimal temperature in the presence or absence of cryoprotectants. Classical freezing includes the successive steps such as pregrowth of samples, cryoprotection, slow cooling (0.5-2ºC / min) to a determined prefreezing temperature (usually around -40ºC, rapid immersion of samples in liquid nitrogen (LN), storage, rapid thawing and recovery. It needs technical skill as it has several steps such as freezing, storage, thawing, re-culture of living plant cells during cryopreservation against cryogenic injuries. Maintenance of cryogenic cultures in LN at -196°C or in the vapor phase of LN at -135°C is in such a way that the viability of stored tissues is retained following re-warming. Various cryopreservation methods are being used for various plant species such as vitrification, encapsulation–dehydration and encapsulation vitrification (Stacey & Day, 2007). The genetic stability can be maintained during cryopreservation that has been proved by molecular marker study (Liu et al., 2008). The cryopreserved tissue has considered as safer, clean, disease free genetic stock for international exchange (Feng et al., 2011).

6. *In Vitro* Gene Bank

The maximum possible genetic diversity of the particular genetic stock can be maintained using *in vitro* gene bank technique. The International Plant Genetic Resources Institute (IPGRI) as well as the Consultative Group on International Agricultural Research (CGIAR) Centers like the International Center for Agricultural Research in the Dry Areas (ICARDA) is heavily involved in the conservation of rare and endangered plant species by maintaining in *vitro gene* bank (Reed et al., 2004; ICARDA, 2014). Several DNA marker-based techniques are generally useful methods in monitoring variability in rare and endangered plant species. Another important fact is the simple and uniformly available facility in the germplasm conservation program is very important factor in monitoring gene bank at the organizational level. *In vitro* derived genetic variation in somaclones are of important source of genetic variability if it persists generation after generation as genetic variant. A large number of biotechnological approaches can be of useful technique in determining the genetic variability among the germplasms. The variability can be determined either using biomolecular markers or by using DNA based markers such as restriction fragment length polymorphism (RFLP), random amplified polymorphic DNA (RAPD), amplified fragment length polymorphism (AFLP), sequence characterized amplified regions (SCAR), simple sequence repeat (SSR), inter simple sequence repeat (ISSR), those are either PCR based or non-PCR based techniques (Glaszmann et al., 1987; Khan et al., 2012). Determination of storage conditions, provision of inventory, and evaluation of viability and verification of genetic stability are very important components of gene bank structure. DNA banking can be considered as a means of complimentary method for the conservation of plant species together with conventional *ex situ* approaches in preserving biodiversity.

7. Transgenic crops and farm animals

Production of transgenic crops and animals is another application of biotechnology in biodiversity conservation. Transgenic crops are more likely to increase agricultural biodiversity and help maintain native biodiversity rather than to endanger it. Such crops may prove to be very useful to the farmers and can be of commercial value. However, the practical benefits and risks of the crops need to be assayed in the field and their products scrutinized.

In case of animals, several lines of transgenic farm animals have been produced, but none have been commercialized. Some lines are made for the pharmaceutical industry to produce drugs in their milk. Others may show improved resistances towards certain infections. Biotechnological methods have many advantages to conventional captive breeding procedures.

Since the animals need not to be moved around, less stress is experienced and the problem of space for keeping the animals is also solved since samples can be taken in the wild. Storage of genetic resources will help to preserve biodiversity and counter the effect of genetic drift on small populations. Even if an animal dies, its genes will still be available for future breeding work.

CONCLUSION

The loss of biodiversity is a great threat to the survival of mankind and the ecosystem at large. Even though several strategies have been adopted in understanding and conserving plant and animal diversity throughout the world, both *ex situ* and *in situ* methods of biodiversity conservation are equally important. Though the biotechnological methods of biodiversity conservation offer many advantages to conventional procedures but these technologies have various social consequences. Biotechnology applications must be integrated with ongoing conventional breeding and development programs in order to succeed. Additionally, the generation, adaptation, and adoption of biotechnologies require a consistent level of financial and human resources and appropriate policies need to be in place.

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