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Recent Advances of Tissue Culture in Crop Improvement

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Plant-regeneration techniques in tissue culture have been used in many fields, including gene-function research, transgenic breeding, and rapid micro propagation. In gene-function research, multiple methods, including over expression, gene knockout, and genome editing, rely on genetic transformation in plants.

Tissue culture is that the in vitro aseptic culture of cells, tissues, organs or whole plant under controlled nutritional and environmental conditions1 often to supply the clones of plants. The resultant clones are true-to sort of the chosen genotype. The controlled conditions provide the culture an environment conducive for his or her growth and multiplication. For instance, in strawberry millions of plants can be produced from 1 mm explant in one year now scores of multimillion dollar industries around the world propagate a variety of plant species through tissue culture.

Plant tissue culture technology is being widely used for giant scale plant multiplication. Plant tissue culture methods offer a rich scope for the creation, conservation and utilization of genetic variability for the improvement of field, vegetable and horticultural crops. Micropropagation of selected ornamentals, fruit and forest plants species, is one of the best and most successful examples of the commercial application of tissue culture technology. This technique has great potential for rapid, large scale and true-to-type multiplication. Small pieces of tissue (named explants) are often wont to produce hundreds and metabolites. Small pieces of tissue (named explants) are often wont to produce hundreds and thousands of plants during a continuous process. Micropropagation starts with the choice of plant tissues (explants) from a healthy, vigorous mother plant. Any a part of the plant (leaf, apical meristem, bud and root) can be used as explants. In India, the micropropagation protocols for sugarcane, potato, strawberry, mentha, gladiolus, carnation and chrysanthemum which are now being commercially exploited in the state for rapid mass multiplication of these plants.

1. Explants are often multiplied into several thousand plants in relatively short period of time and space under controlled conditions, regardless of the season and weather on a year-round basis

2. Endangered, threatened and rare species have successfully been grown and conserved by micropropagation due to high coefficient of multiplication and little demands on number of initial plants and space.

Tissue culture technology offer environmentally friendly industries to flourish. There are more than 200 laboratories in the World including about 50 in India, and 7 in Punjab and Chandigarh each producing more than 1.5 million plants per annum. USA is the largest producer followed by Asia, The Netherlands, other European countries, Australia and New Zealand, France, Italy and Israel. It is estimated that more than 500 million plants belonging to different plant species are being produced through micropropagation, annually in the different parts of the world. The clean planting material can certainly improve the yield potentials of our vegetative propagated crops like sugarcane, potato, strawberry, mint, sweet potato, banana and tapioca. It is likely that automation of multiplication systems will be commercially feasible within the next few years for several species including potato micro tubers, lily bullets and gladiolus corms. Improvement of somatic embryogenesis, coupled with embryo desiccation and encapsulation technology may lead to the utilization of artificial seeds for mass cloning of plants (Redenbaugh *et al*., 1988). Soma clonal variation - the variation among the callus derived plants is a potent emerging aspect for broadening the genetic base and thus obtaining incremental improvement in the commercial cultivars, more particularly, in the vegetatively propagated species.(Evans and Sharp, 1986). Using the technique of in vitro selection many

million cells/protoplasts (cell without cell wall can be screened against various biotic and abiotic stress factors in a single Petri dish which is more efficient as compared to the screening of similar number of plants in the field which required more time and space as well. Several interesting and potentially useful traits have been recovered using this method in sugarcane, potato, tomato, corn, rapeseed and mustard, rice and alfalfa. However, under several situations, the lower plant regeneration ability and the lack of correspondence in expression of the trait in vitro and in plants are the major problems. We have induced somaclonal variation in sugarcane and potato. Selected somaclones are under field evaluation.

Production of haploids through bulbosum, anther/pollen culture methods, has been exploited for the early release of varieties (Foroughi, Wehr and Weinzel, 1989). For instance, production of haploids/doubles haploids through another culture from F_1 plants, results in true breeding plants in less than one year, which are otherwise obtained after 7-8 generations through conventional methods. In rice, the anther culture breeding has been and would be highly rewarding in early release of new varieties. The largest number of cultivars has come from barley, using `bulbosum' maternal haploid method. Seven barley varieties have been released in Canada alone. Besides, using the technique of another culture haploids have been produced in more than 50 genera. Several cultivars are either in test or have been released in rice, wheat, maize, rapeseed and mustard in China, Canada, Denmark, USA and France. But in many instances, the poor androgenesis, occurrence of mixoploids and albino plants have been the recurring problems. Using anther culture, we have developed a population of doubled haploids from an elite cross involving high yielding rice x super rice. Besides wheat x maize crosses are being exploited for production of wheat haploids.

Since the possibility of producing useful secondary products in plant cell culture was first recognized in 1970s, considerable progress has been made and a number of plant species have been found to produce secondary products such as shikonin, diosgenin, caffeine, gutathion and anthraquinone. Large scale production of such compounds in increasingly becoming popular with the industry where some physical and chemical conditions for growth and product formation have been optimized.

Embryo culture is the practical approach to obtain interspecific and intergeneric hybrids among otherwise hard to cross parents (Gosal and Bajaj, 1983). It has been successfully used to transfer desirable genes from wild relatives into cultivated varieties of several field and vegetable crops. Somatic cell hybridization involving fusion of protoplasts from different species is considered an important approach to combine characteristics even from otherwise sexually incompatible species and to obtain cybrids (cytoplasmic hybrids) and organella recombination, not possible through conventional methods (Hinnisdaels *et al.*, 1988). Earlier efforts were to combine full genomes from both the parents and develop symmetric hybrids. However, the somatic hybrids thus produced, particularly among the phylogenetically remote species, have exhibited somatic incompatibility, genetic instability and sterility. It is, now obvious that these monsters cannot be incorporated into the breeding programmes. Therefore, the interest has moved from creation of novel hybrids to production of cybrids, chromosome transfer and gene introgression.

As a consequence, some useful agronomic traits like male sterility, herbicide and disease resistance have been transferred from wild species into cultivated varieties of potato, tomato and brassica. Furthermore, the development of gene transfer systems have now greatly expanded the gene sources which may come from wild relatives, unrelated plants, bacteria, viruses, fungi, animals or even from chemical synthesis in the laboratory. Using various vector systems (Agrobacterium and viruses) and vectorless systems (liposome's, microinjection, laser micro beam and particle gun), the transgenic plants has been produced in more than 100 plant species. It has been demonstrated that biotechnology can produce plants that contain their own pesticide and resist lethal doses of weed-killers and plant diseases (Dale *et al.*, 1993). Transgenic plants, carrying agronomic ally useful genes such as herbicide resistant, virus resistant and

insect resistant of several field crops are in field trials in many countries and commercial releases have also been made in tomato, cotton corn etc. We have developed Bt basmati and the transgenics are under glass house evaluation.

Current and future status of plant tissue culture

The past decades of plant cell biotechnology has evolved as a replacement era within the field of biotechnology that specializes in the assembly of an outsized number of secondary plant products. During the last half of the last century the event of gene-splicing and molecular biology techniques allowed the looks of improved and new agricultural products which have occupied an increasing demand within the productive systems of several countries worldwide (James C 2008). Nevertheless, these would are impossible without the event of tissue culture techniques, which provided the tools for the introduction of genetic information into plant cells (Pareek 2005). Transgenic plants represent uneconomical alternative to fermentation-based production systems. Plant-made vaccines or antibodies (plantibodies) are especially striking, as plants are freed from human diseases, thus reducing screening costs for viruses and bacteria toxins. The amount of farmers who have incorporated transgenic plants into their production systems in 2008 was 13.3 million, in comparison to 11 million in 200734.

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