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HOW BIOTECHNOLOGY WILL BRING THE NEXT AGRICULTURAL REVOLUTION AND RECENT DEVELOPMENTS IN THE FIELD

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Agriculture has come a long way, from simply harvesting wild strains of cereals and domesticating them to commercial cultivation of genetically modified plants. One of the most significant events in the development of modern agriculture was the green revolution (1940s–1960s), during which the overall yield of crops was drastically increased by the introduction of high-yielding varieties and the application of chemical fertilizers and pesticides. After the green revolution, it seemed that food production could keep pace with worldwide population growth.

But now the ever-rising human population is facing the same threat of food crisis again. Due to extensive use of chemical fertilizer and high-yielding varieties, we have created new problems for us and the environment, Loss of soil fertility, erosion of soil, soil toxicity, diminishing water resources, pollution of underground water, salinity of underground water, increased incidence of human and livestock diseases and global warming, Extinction of Indigenous Varieties of Crops, are some of them. Therefore, we have to find new and sustainable ways of enhancing food production for the growing population, while also taking into account the current climate change situation and environmental issues our world is experiencing.

The green revolution during the 1940s relied mainly on plant breeding and crossing to develop high-yielding varieties. However, using only those conventional breeding techniques will not be sufficient to maintain the food supply at the current population growth rate. In the last few decades plant biotechnology and genetic engineering have acted as a very useful tool to supplement the drawbacks of conventional plant breeding. In both traditional and genetically engineered plant types, biotechnology tools assist in the production of better

kinds. Not only that, the use of biotechnological tools has helped in the detection and diagnosis of plant diseases, thus assisting in crop protection.

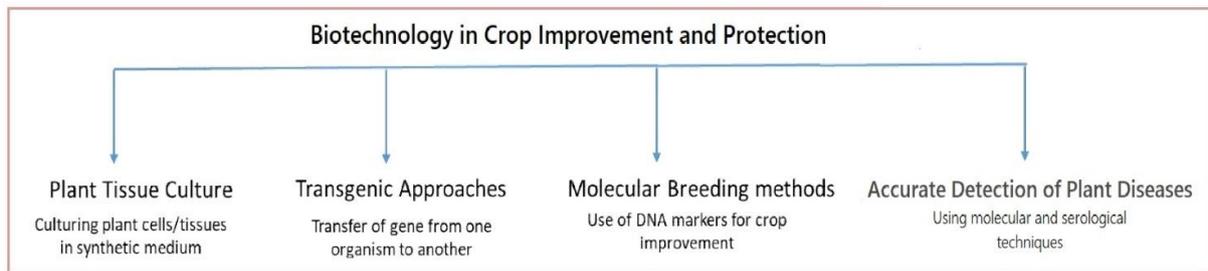


Fig 1: Approaches of Biotechnology in Agriculture

Biotechnological Approaches in Agriculture

Agricultural biotechnology refers to the use of biological organisms or a range of tools for the improvement of plants, animals, microorganisms, or food derived from them. The application of biotechnology in agriculture has benefitted farmers, producers, and consumers. Following are some biotechnology tools used in agriculture:

A. Plant Tissue Culture

Plant tissue culture, in its broadest sense, refers to the *in vitro* growing of live plant cells, tissues, or organs (seeds, embryos, single cells, protoplasts) on a nutritional medium in an aseptic environment. Plant tissue culture techniques include micropropagation, somatic embryogenesis, somaclonal variation, meristem culture, anther culture, embryo culture, protoplast culture, cryopreservation, and secondary metabolite generation.

B. Transgenic Approaches/Genetic Engineering

Biotechnology is widely employed in the production of genetically modified (GM) crops, in which one or more genes coding for desired features have been added through the genetic engineering process (GE). The gene used to create the transgenic might come from the same species or species and organisms unrelated to the recipient organism. Transgenic technology is the method of transferring genes from related or unrelated species to desired agricultural plant species for genetic study and direct DNA modification. This gene technology is also known as recombinant DNA technology or genetic engineering.

Mainly there are two techniques for introducing foreign genes into plants. The first technique employs *Agrobacterium tumefaciens*, a soil-borne, Gram-negative bacterium that

causes crown gall disease in several species. This bacterium possesses a plasmid containing tumor-inducing genes (T-DNA) as well as other genes that aid in T-DNA integration into the host genome. This is accomplished by deleting the majority of the T-DNA but retaining the border sequences (24 bp) that integrate a foreign gene into the genome of cultivated plant cells. The second approach is a "gene gun," which bombards plant cells with gold particles containing foreign DNA. Some of these particles get past the plant cell wall and into the cell nucleus, where they cause damage to the DNA.

Over the last 15 years, the combination of recombinant DNA technology with tissue-culture techniques has resulted in the effective transformation and generation of transgenic in a wide range of agricultural plants.

C. Marker Assisted Selection

Marker-assisted selection (MAS), where DNA markers are used to guide, support, and streamline plant breeding efforts. Molecular marker-aided genetic analysis aids in gene identification by studying DNA sequences to identify genes, QTL (Quantitative trait loci), and molecular markers; and to correlate them with the organism. Molecular marker-aided selection aids in the identification and tracking of previously recognized DNA segments across generations. Molecular marker-assisted breeding uses molecular markers, linkage maps, and genomics to modify and improve plant or animal traits based on genotypic assays.

D. Detection of Plant Diseases using Molecular Approaches

Recent improvements in molecular biological methods have improved the identification and diagnosis of new, emerging, previously reported, and re-emerging fungal plant diseases. Polymerase chain reaction (PCR)-based tests, isothermal and post-amplification tools, hybridization methods, and next-generation sequencing (NGS) techniques are well-known for diagnosing phytofungus diseases. These molecular-based techniques have effectively detected and diagnosed symptomatic and asymptomatic diseases caused by culturable and unculturable fungal pathogens in single and co-infections of significant field, horticultural, floricultural, ornamental, and forest plant species. When the sample load is insufficient to detect, quantitative PCR has been widely employed in the quantification and identification of causal organisms.

Recent Developments

Let's now discuss some of the most recent innovations supporting the development of agricultural biotechnology.

- In more recent times, **omics technologies** like as metabolomic and transcriptomic-assisted breeding have been applied in MAS. The future of agricultural biotechnology will heavily rely on developing new techniques to aggregate and evaluate various data kinds in order to optimize the knowledge accessible to breeders.
- **Genomics** is the most powerful approach for interpreting crop species' stress response with adaption features or identifying underlying genes, alleles, or quantitative trait loci.
- Recent breakthroughs in genotyping, sequencing, and phenotyping platforms (phenomics) have turned molecular breeding into **genomics-aided breeding (GAB)**. Marker-aided selection (MAS) and genomic selection are the most often employed methodologies for genomics-assisted breeding (GS). SNP arrays, which are very inexpensive and automatically genotyping assays, are being used for high throughput genotyping. It is commonly utilized in crop genetic investigations, including as genome-wide association studies (GWAS), linkage map building, genomic selection, population structure analysis, and gene mapping.
- **Site-directed nucleases (SDN) for targeted gene insertion or replacement-** Agrobacterium tumefaciens-mediated and DNA-coated particle bombardment transformation technologies have a few limitations, such as the possibility for endogenous gene disruption or misexpression of neighboring genes via trans- or cis-gene regulation. Another disadvantage of using numerous transgenes is that they would likely integrate into various chromosomes and segregate independently, complicating downstream breeding. To overcome these possible flaws, researchers are looking at using SDNs to precisely introduce DNA elements into the plant genome at specified DNA breaks using the plant's DNA homology-guided repair process. Multiple inserted genes can be integrated at a single genomic safe harbour, a designated chromosomal location with minimum positional effects, to optimize transgenic effectiveness without disrupting essential cell activities.

- **RNA therapeutics: RNAi and antisense-** RNAi (RNA interference) is a gene silencing method that uses double-stranded RNA to suppress protein synthesis in target cells. Antisense technology achieves the same outcome using single-stranded RNA. Antisense technology has produced promising results in the development of FlavrSavr, a tomato cultivar with improved shelf-life.
- **Clustered Regularly Interspaced Short Palindromic Repeats (CRISPR)/Cas9** (CRISPR-associated protein 9) genome editing technology has shown considerable promise in tackling new agricultural difficulties fast. Recently, Haque et al. showed the potential of CRISPR/Cas9 for improving crop resilience against new pests and abiotic challenges in tropical regions. It is capable of accurately altering the genome sequence of any creature, including plants, to obtain the desired characteristic. Several techniques, such as optimizing promoters to drive and express Cas9 and utilizing various fluorescence reporters and selection markers, have recently been investigated to enhance plant transformation by CRISPR/Cas9. The CRISPR/Cas gene-editing method may produce heritable, targeted alterations while simultaneously addressing concerns about the presence of foreign DNA sequences by producing transgene-free plants.
- **Loop-mediated amplification (LAMP)** a modification of PCR is currently showing success in the detection of fungal diseases and has aided in the identification of *Alternaria* spp., *Colletotrichum* spp., *Fusarium* spp., *Verticillium* spp., *Puccinia* spp., *Botrytis* spp., etc. NGS may be used to find novel and emerging diseases by sequencing fungal genomes on several platforms with no prior knowledge of the pathogen's sequence.

Conclusion

From the green revolution to the gene revolution, agriculture has come a long way. Every day, it evolves at a breakneck pace. With the capacity to understand and manipulate the genetic composition of organisms using biotechnological methods, we can meet the rising demand for food by developing novel crop varieties with a higher yield, improved resilience to biotic and abiotic stresses, and environmental sustainability. The application of biotechnology in agriculture has not only increased crop yield but also lowered production costs by reducing the necessity for inputs (pesticides) and improved farmers' lives.

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