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Farming the Future: Advanced Techniques in Modern Agriculture Volume II

Editors:

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Volume II

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PREFACE

Welcome to "Farming the Future: Advanced Techniques in Modern Agriculture" This book represents a journey into the transformative landscape of contemporary farming practices. As our world grapples with burgeoning populations, shifting climates, and environmental challenges, the need for sustainable and efficient agricultural methods has never been more pressing.

In these pages, we explore the cutting-edge techniques and technologies that are reshaping the agricultural sector. From precision farming and hydroponics to genetic engineering and AI-driven analytics, each chapter delves into innovations that promise to enhance yields, conserve resources, and mitigate environmental impact.

The evolution of agriculture is not merely a matter of technological advancement but a profound shift in how we steward the Earth's resources responsibly for future generations. It is about harnessing innovation to ensure food security without compromising the integrity of our ecosystems.

Throughout this book, experts and pioneers in the field share their insights, challenges, and successes, offering a comprehensive guide to those who are passionate about shaping the future of farming. Whether you are a seasoned agriculturalist, a researcher, a policymaker, or simply curious about the future of food production, "Farming the Future" aims to inform, inspire, and catalyze meaningful change.

As we embark on this exploration together, let us envision a future where agriculture not only sustains but thrives—a future where innovation and sustainability go hand in hand to feed a growing planet while safeguarding its natural resources.

Thank you for joining us on this journey into the heart of modern agriculture.

Editors

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ALIEN GENE INTROGRESSION APPROACH FOR CROP IMPROVEMENT

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Abstract:

Alien genes have contributed several traits in the crop plants which are not available in the cultivated background. These have helped plant breeders in creating newer genetic diversity, thereby providing additional avenues of selection of better plant types. Vertical and horizontal transfer of alien genes has changed the fate of several crops by imparting resistance to diseases and insect-pests, tolerance to abiotic stresses. Several challenges such as pre and post fertilization barriers in distant crosses, problems in normal chromosome pairing, linkage drag, pleiotropic effects and role of recipient genome background on the expression of introgressed alien gene(s) and in HGT (Horizontal gene transfer), erratic regeneration protocols, difficulty in isolation of genes from wild species and their expression in recipient plants, and possibilities of gene flow pose significant challenges to make alien gene transfer a routine process across all crop species. However, recent advances in sequencing and genotyping technologies have made it possible to develop molecular markers as well as undertake genotyping at large scale in both major as well as minor (or so called orphan crop species) that can be used not only for developing high density genetic and physical maps but also for generating transcriptome or sequence data. In parallel, the high-throughput sequencing and genotyping approaches can be used to detect genetic variation existed in germplasm collection not only in cultivated gene pool but also in landraces and wild species. Furthermore, the QTL (Quantitative trait locus) or genes or superior alleles for the trait of interest identified through linkage mapping, association mapping, AB-QTL approach can be introgressed or pyramided in elite varieties or genotype of interest by using MAGIC (Multi-parent advanced generation inter-cross), MABC (Marker-Assisted Backcrossing), MARS (Marker-Assisted Recurrent Selection) or GWS (Genome-Wide Association Studies) approaches. Alien gene introgression is a critical tool in crop improvement, addressing the multifaceted challenges facing agricultural and food production. It enable the development of crops that are more resilient, productive and adaptable contributing to global food security, sustainability and economic growth.

Keywords: Alien gene, HGT, VGT, Molecular Markers, QTL.

Introduction:

The twentieth century has witnessed tremendous improvement in global crop production. Besides several factors such as increase in cultivated area, improved agronomic practices, increased use of plant protection measures and better crop management, improved varieties of

crop plants have played a dramatic role in improving the productivity of different crops. The genetically improved crop cultivars have been developed through modern plant breeding by introducing improved alleles at existing loci through conventional hybridization, of late aided by molecular marker technology and genetic transformation.

The aim of all these techniques has been either exchange of genes between sympatric or neighboring populations of crops and related taxa or transfer of genes from related taxa into the cultivated gene pool of a crop. This led to the development of numerous improved cultivars with high yield, stress resistance and superior agronomic performance. In nature, gene transfer from one population to another is slow as compared to man-made systems, whereas it is faster and often mediated by hybridization followed by a number of back crossings and rigorous phenotypic selections.

Alien gene introgression is a breeding technique used in crop development to introduce specific desirable traits from wild or closely related species into cultivated crop plants. This approach is employed to enhance the genetic diversity of crops, improve their resistance to pests, diseases, and environmental stresses, and increase their overall productivity and quality. Here's an overview of the alien gene introgression approach for crop development:

(a) Identification of Target Traits: The first step in this approach is to identify the specific traits or characteristics that need to be introduced or improved in the crop. These traits could include disease resistance, tolerance to environmental stresses (e.g., drought, heat, cold), increased yield, enhanced nutritional content, or other desirable features.

(b) Selection of Donor Species: Once the target traits are identified, the next step is to select a donor species or related plant that possesses these traits. Donor species are often wild relatives of the crop or sometimes other cultivated varieties with the desired traits.

(c) Cross Breeding: Cross breeding is done to create hybrid plants that carry genetic material from both the crop and the donor species. This is typically achieved through controlled pollination and hybridization techniques. The hybrid plants are referred to as F1 hybrids.

(d) Backcrossing: The F1 hybrid plants are then backcrossed with the original crop plant. This process involves repeatedly crossing the hybrid with the cultivated crop for several generations (F₂, F₃, etc.) to gradually increase the proportion of crop genes while retaining the desired alien genes.

(e) Selection and Marker-Assisted Breeding: During backcrossing, selection is performed to choose plants that exhibit the target traits and have a higher proportion of crop genes. Marker-assisted breeding can be used to identify and track the presence of specific genes associated with the desired traits.

(f) Genetic Screening: Molecular techniques such as DNA markers can help in the identification and tracking of specific alien genes in the backcrossed plants.

(g) Field Testing: Promising backcrossed plants are subjected to field trials to assess their performance under real environmental conditions. This step helps confirm that the desired traits are stably integrated into the crop and that there are no unexpected negative side effects.

(h) Regulatory Approvals: Before a genetically improved crop can be commercialized, it must go through regulatory processes to ensure it is safe for consumption and the environment. These processes vary by country and region.

(i) Release and Adoption: Once a new crop variety is approved and deemed safe, it can be released to farmers for cultivation. Farmers can then adopt these improved varieties to benefit from their enhanced traits.

Alien gene introgression is a valuable tool in crop breeding because it allows for the transfer of genetic diversity and desirable traits from related species, helping to address challenges in modern agriculture, such as pest resistance and climate change adaptation. However, it is essential to carefully manage the introgression process to avoid unintended consequences and maintain the integrity of the crop being improved.

The influence of alien gene transfer on the way the fate of a crop can be changed was first demonstrated as early as in 1956 when a small segment from *Aegilops umbellulata* Zhuk carrying a gene for resistance to leaf rust was translocated onto the wheat chromosome 6B (Sears, 1956). This revolutionary work ushered an era of utilization of wild genetic resources for the improvement of crop plants. Consequently, interspecific and intergeneric hybridization have been widely adopted by plant breeders across different crops and used to develop improved cultivars with enhanced agronomic performance, resistance to biotic and abiotic stresses, and quality improvement. However, the success in utilizing wild genetic resources for transfer of desirable genes has not been uniform across all crop species since alien gene transfer largely depends upon availability of such resources, ease of hybridization and expression of the trait of interest in the progeny. Owing to these requirements, routine alien gene introgression from wild species is still a challenge before scientists for harnessing the desirable genes of wild species across all species. While evaluation of wild relatives and identification of genes conferring traits of interest themselves pose greatest challenges to breeders, effecting successful hybridization and obtaining a viable progeny with the gene(s) of interest transferred into them is further a difficult task. For involving wild species in hybridization, these need to be available in the vicinity of the recipient parent and their flowering must synchronize with that of the cultivated species. Raising wild species and exotic germplasm in field condition is often not easy and development of controlled conditions such as glass houses and plant growth chambers requires huge investments on time and money.

Alien gene introgression is sometimes associated with several other difficulties such as linkage drag and pleiotropic effects; while in some instances this is associated with some unforeseen advantages also such as development of chromosome elimination technique for doubled haploidy breeding in wheat and barley. Advancements of in vitro techniques, in vivo and in vitro hormonal manipulations, techniques such as somatic hybridization and protoplast fusion and the most recent development of cisgenesis and intragenesis have offered commendable opportunities towards alien gene introgression.

Need for Alien Gene Transfer: Genetic variation is essential for developing new plant varieties and this can be created by introducing genes from a related species, sometimes from a relatively distant species or even an unrelated species. The need for gene transfer in a crop species depends upon the extant genetic variability in that crop as well as availability of a trait of interest in the donor in intense form. In most of the cultivated crop species, limited popular and high yielding varieties are grown over wide areas and these are often derived from a relatively narrow representation of gene pool, mostly from the primary gene pool, and therefore these have a narrow genetic base and limited genetic buffer. Making selections for desired traits such as non-shattering habit, uniform maturity, improved seed fertility, seed dormancy, increased seed number, increase in seed and fruit size, modified plant architecture and conversion from perennial to annual forms during the process of crop domestication led to a gradual loss in genetic diversity (Tanksley and McCouch, 1997).

This reduction or loss in genetic diversity during crop domestication could be attributed to (i) selection by human beings for desirable “domestication related traits” (ii) genetic drift in the form of “domestication bottlenecks” and (iii) modern plant breeding practices that resulted in the development of high yielding and uniform crop varieties. This reduction in diversity has been more prominent in self-pollinated crops like wheat where the level of genetic variation in cultivated pool has often been reported to drop below 5 % of that available in nature. It makes crops more vulnerable to biotic and abiotic stresses. This may also result in huge losses in yield and quality as observed previously by the attack of shoot fly and Karnal bunt in India and the Southern corn leaf blight in the United States (Tanksley and McCouch, 1997). Moreover, it reduces chances to identify new and useful gene combinations for crop improvement. In this way, modern plant breeding although increased crop productivity worldwide, it also eroded the genetic variability of the crops. Consequently, our major crop species represent the relatively few species that were selected by our ancestors from a multitude of extant species, and the resulting narrow germplasm forms the basis of modern monoculture in many areas of the world. This makes them fragile to global climate change and vulnerable to new races of pathogens and insect pests. Due to narrow genetic variability, options to execute selection for desirable plant types also become limited. To overcome these concerns and for further genetic improvement in crops plants, the natural variation available in wild relatives, landraces, and primitive cultivars of the crop species is required to be harnessed for a rapid and sustainable improvement of crop species for many years (Tanksley and McCouch, 1997).

Wild species are a rich reservoir of useful alien genes which are no longer available within the cultivated gene pool (Tanksley and McCouch, 1997). Since these species have had much longer time and increased opportunities to evolve and adapt to natural environments, therefore, these often have genes for resistance to diseases and insect pests and for tolerance to drought, temperature stress, salinity and other extreme environmental conditions. Further, they have wide genetic buffers to withstand unexpected adversities.

Sources of alien genes and their characterization: In crop breeding, alien genes can be sourced from wild or closely related plant species to introduce specific traits or characteristics into

cultivated crops. The characterization of these alien genes is crucial to ensure that they are effectively integrated into the crop's genome and do not have any unintended negative effects. Here are some sources of alien genes and their characterization:

(a) Wild Relatives of Crops: Wild relatives of cultivated plants often harbor a reservoir of genetic diversity and traits that can be beneficial for crop improvement. These species are closely related to the crop of interest and may have evolved specific adaptations or resistances to pests, diseases, or environmental stresses.

(b) Other Cultivated Varieties: In some cases, alien genes can be sourced from other cultivated varieties of the same or closely related crops. These varieties may have specific traits that are not present in the target crop but are desired for improvement.

(c) Cross-Compatible Species: In some cases, alien genes can be obtained from species that are cross-compatible with the target crop. These species may belong to the same genus or family and can be hybridized with the crop to transfer desired traits.

CHARACTERIZATION OF ALIEN GENES:

(a) Phenotypic Characterization: The first step in characterizing alien genes involves assessing the phenotypic traits associated with these genes. This can include evaluating the appearance, growth habits, resistance to pests and diseases, and tolerance to environmental stresses of the donor species or varieties.

(b) Molecular Characterization: Molecular techniques are used to identify and characterize the specific genes or DNA sequences responsible for the desirable traits. This can involve DNA sequencing, gene mapping, and marker-assisted breeding.

(c) Genetic Mapping: Genetic mapping is used to determine the location of the alien genes within the genome of the donor species and the recipient crop. This information is essential for tracking the introgression of these genes during breeding.

(d) Linkage Analysis: Linkage analysis helps determine if the alien genes are tightly linked to other genes in the donor genome. Understanding the linkage can be crucial to avoid inadvertently introducing unwanted traits.

(e) Functional Analysis: Functional analysis involves determining how the alien genes function in the context of the target crop. This includes understanding the biochemical and physiological mechanisms underlying the desired traits.

(f) Expression Patterns: Analyzing the expression patterns of alien genes in the target crop can help ensure that the introduced genes are active and functioning as intended. This can be done through techniques such as gene expression profiling.

(g) Phenotypic Evaluation of introgressed lines: After introgressing the alien genes into the crop, the resulting lines should be carefully evaluated in field trials and controlled environments. This allows for the assessment of how well the traits are expressed and whether any unintended side effects are present.

Regulatory Compliance: For crops intended for commercial release, regulatory agencies often require comprehensive characterization of the introduced genes to ensure the safety of the modified crop for human consumption and the environment. Characterization of alien genes is an

ongoing process that combines traditional breeding techniques with modern molecular and genetic tools. This helps ensure that the introduced genes are stable, functional, and do not result in any unexpected consequences in the cultivated crop.

The genes present in distant relatives (i.e., wild species) are usually known as alien genes. Therefore, these species are important source of such alien genes in crop plants. Among these species, phylogenetic relationships have been established on the basis of cross ability of cultivated species with the wild species and other cytological and molecular analysis. This has led to characterization of wild species into primary, secondary, and tertiary gene pools according to the gene pool concept of Harlan and De Wet (1971). This gene pool concept provides the knowledge regarding the possibilities of transferring alien genes controlling desirable traits from wild species either through conventional crossing or by using the advanced modern technologies. In general, species within the primary gene pool are easily crossable with each other and hence have been used easily for transfer of alien genes. Although wild species belonging to secondary gene pool may also cross readily with cultivated species, some post-zygotic barriers restrict their use in alien gene transfer. However, recent advances in tissue culture techniques have made it feasible to use the species of this group. Wild species of tertiary gene pool are not found cross compatible with cultivated species. A large proportion of wild species belongs to this group and consequently is of no use for crop improvement through sexual manipulations.

Importance of Alien Gene Introgression: Genetic variation of crop plants is continuously decreasing due to domestication and modern plant breeding practices. This has although resulted in development of high yielding, uniform crop varieties, but it has happened largely at the cost of extinction of primitive ancestors. Consequently, these uniform and high yielding varieties become more vulnerable to attacks by diseases and insect-pests leading to heavy losses and some cases, to near extinction of a crop.

Alien gene introgression is an important and valuable technique in crop breeding and genetic improvement for several reasons:

(a) Genetic Diversity: Alien gene introgression increases the genetic diversity of crop plants by introducing genes from wild or related species. This genetic diversity is crucial for enhancing a crop's ability to adapt to changing environmental conditions, resist pests and diseases, and respond to evolving agricultural challenges.

(b) Trait Improvement: By incorporating genes from donor species, crops can acquire desirable traits that may not be present in their current genetic makeup. These traits can include disease resistance, tolerance to abiotic stresses (e.g., drought, heat, cold) increased yield, improved nutritional content, and enhanced quality characteristics.

(c) Pest and Disease Resistance: Alien gene introgression can confer resistance to specific pests and diseases that threaten crop production. This reduces the need for chemical pesticides and can lead to more sustainable and environmentally friendly farming practices.

(d) Abiotic Stress Tolerance: In the face of climate change and increasing environmental stressors, such as drought and extreme temperatures, alien genes can provide crops with the

resilience needed to thrive under adverse conditions. This is crucial for ensuring food security and maintaining agricultural productivity.

(e) Reduced Reliance on Chemical Inputs: Crops with improved resistance traits acquired through alien gene introgression may require fewer chemical inputs, leading to reduced environmental impact and lower production costs for farmers.

(f) Improved Nutritional Content: Alien genes can be used to enhance the nutritional content of crops. This is particularly important for addressing malnutrition and health-related issues, as well as increasing the value of agricultural products.

(g) Sustainable Agriculture: Alien gene introgression can contribute to sustainable agriculture practices by reducing crop losses, conserving natural resources, and enhancing overall agricultural productivity.

(h) Crop Variety Development: The technique enables the development of new crop varieties with diverse characteristics, allowing farmers to choose varieties that are best suited to their specific local conditions and preferences.

(i) Economic Benefits: Improved crop traits resulting from alien gene introgression can lead to increased crop yields, higher market value, and improved income for farmers. This, in turn, contributes to food security and economic development in rural areas.

(j) Reduction of Post-Harvest Losses: Traits introduced through alien gene introgression, such as resistance to storage pests and diseases, can help reduce post-harvest losses, ensuring more of the harvested crop reaches consumers.

(k) Adaptation to Global Challenges: Alien gene introgression is essential for adapting crops to the ongoing challenges of a changing climate, population growth, and evolving agricultural practices.

(l) Biodiversity Conservation: By preserving and utilizing genes from wild and related species, alien gene introgression contributes to the conservation of biodiversity. This is particularly important for preserving valuable genetic resources.

Alien gene introgression is a critical tool in crop improvement, addressing the multifaceted challenges facing agriculture and food production. It enables the development of crops that are more resilient, productive, and adaptable, contributing to global food security, sustainability, and economic growth.

E.g. Grassy Stunt Virus (GSV) epidemics in Rice: During the early 1970s, before the release of resistant rice cultivars in 1974, GSV epidemics destroyed more than 116,000 ha (287,000 acres) of rice in Indonesia, India, Sri-Lanka, Vietnam, and Philippines. After this, screening of ~17,000 cultivated and wild rice lines for resistance to GSV for 4 years led to the identification of a population of *Oryza nivara* growing wild near Gonda in Uttar Pradesh, India and showing resistant to GSV. This resistance in *O. nivara* for “grassy-stunt virus strain 1” was governed by a single resistance gene. This gene was transferred from *O. nivara* into cultivated rice and it is believed that GSV resistant hybrids containing the wild Indian gene are grown across 110,000 km² of Asian rice fields. This is one of the most important examples showing how wild relatives

of crop plants came to the rescue of cultivated crops and thus prevented massive crop failure and famine.

Challenges in Alien Gene Transfer: Alien gene introgression has opened new ways and opportunities in creating additional genetic variability and providing newer avenues of useful selection in crop plants besides helping in evolution of the crop species. While developments in hybridization strategies and advancement of *in vitro* techniques have made alien gene introgression in cultivated species easier, certain challenges are still there which make alien gene introgression a routine practice a bit difficult in plant breeding. There are two ways to transfer the alien gene(s) into cultivated species: transferring alien gene from cross-compatible wild species through hybridization (vertical gene transfer) and transfer of gene(s) from other sources as well as cross-incompatible wild species through genetic transformation and somatic hybridization (horizontal gene transfer).

(1) Vertical Gene Transfer

Crossability Barriers: Wild species are an important reservoir of useful genes. However their use, especially of those species belonging to the tertiary gene pool, has been limited for transferring the useful genes due to cross ability inhibition and limited recombination between chromosomes of wild and cultivated species (Brar and Khush 1986, Khush and Brar 1992 and Sitch 1990). These cross ability barriers developed during the process of speciation frustrate breeders' efforts in successful hybridization between species of different gene pools. The pre-fertilization cross incompatibility between parent species arises when pollen grains do not germinate, the pollen tube does not reach ovary, or the male gametes do not fuse with female gametes (Chowdhury 1983 and Shanmungam *et al.*, 1983).

Chromosome Pairing: Pairing of chromosomes of wild species with the cultivated species in their hybrids is the key to transfer of gene(s) across species. Genetic control of pairing of chromosomes derived from two different genomes has been identified in wheat, where this gene is known as Ph1. The suppressing of the Ph1 pairing regulation of polyploidy wheats and oat has resulted in desired chromosome pairing and hence alien gene transfers into these crop species. Such cytogenetic manipulations, including the suppression of the Ph1 system, for recombining desirable alien chromatin into wheat were termed as chromosome engineering (Sears, 1972). Essentially similar cytogenetic manipulations affecting gene transfer can also be done in hexaploid oats. In rice, very limited chromosome pairing has been observed at metaphase I in F₁ hybrids of cultivated and wild species. Therefore, it has been difficult to transfer the alien genes from wild species to cultivated species (Brar and Khush, 1986).

Linkage Drag: One of biggest challenges in using wild species for introgression of alien genes into cultivated background is the association of undesirable genes with the useful alien genes, known as linkage drag. Its effect is more severe in crops with diploid genetic systems because their genomes are more sensitive to genetic imbalance compared to relatively more buffered polyploid genomes. This has resulted in exploitation of only a few exotic genes in alien germplasm in agriculture. In wheat, for example, genes other than the targeted gene (e.g., Sr39 transferred from wild species "*Aegilops speltoides* Tauschii") was carried on the alien chromatin

Xu *et al.*, (2008) during introgression, which had a deleterious effect on yield and quality (Latter *et al.*, 1988,). Therefore, it became important to eliminate the excess *Aegilops speltoides* chromatin surrounding Sr39 in order to make this gene useful for fighting against Ug99.

Background Effects: It has been observed that there is a problem of variable expression of introgressed alien genes in cultivated backgrounds. In wheat, Chinese spring and *Leymu sracemosus* translocated chromosomes carrying genes for resistance to Fusarium Head blight have been transferred to different common wheat backgrounds. However, expression of resistant gene was observed to be uniform among the resultant lines. These results demonstrated that effects of genetic backgrounds on the expression of alien resistance genes in wheat were due to epistatic interactions. Therefore, it is evident that efficient manipulation of alien chromatin and selection of proper recipient genotypes play a crucial role in the success of alien introgression for Fusarium head blight resistance.

Pleiotropic effects of Alien Genes: Sometimes introgressed alien genes affect more than one trait. If such effects are positively associated with desirable traits, introgression of alien genes with pleiotropic effects can be useful. However, if these are associated with undesirable traits, it becomes a challenge for breeders to use them in crop. For example in wheat, introgression of leaf rust resistance gene Lr47 (from *Triticum speltoides*) led to an overall reduction of 3.8 % in grain yield; nevertheless it varied significantly across genotypes and environments. At the same time, lines with the alien Lr47segment showed consistent increase in grain and flour protein concentration, while and an increase in flour ash. Similarly, the secondary and tertiary gene pools is still difficult through map-based cloning. Although the wild species are good genetic resources for genes controlling resistance to biotic and abiotic stresses as well as quality traits, these could not be used extensively in breeding programs in either way.

(2) Horizontal Gene Transfer (HGT)

Regeneration Protocol: Horizontal transfer of alien genes across genera requires suitable regeneration system for the development of transgenic plants. This is one of the major challenges, especially in recalcitrant species such as food legumes that restrict transfer of alien genes through genetic transformation (i.e., transgenics). Tissue culture techniques are part of a large group of strategies and range from molecular genetics to recombinant DNA studies, genome characterization, gene-transfer and in vitro regeneration of plants. All these tools require totipotent tissues that readily respond to tissue culture procedures. In most of the species, in vitro regeneration is highly genotype specific and cultivated varieties are rarely amenable to regeneration. Additionally, morphogenesis is generally very slow and very often there are problems like development of albinos and vitreous tissues, and no response in dedifferentiated calli. Therefore, successful and reliable plant regeneration in many crop species still remains an aspiration that requires considerable refinement in technology and training of the human resources to develop the skills that are needed to generate green plants.

Isolation of Genes from Wild Species: Desirable genes present in the background of wild species belonging to primary gene pool have been exploited in development of improved varieties in several crops. These can be isolated through map-based cloning. However

introgressions are accompanied by linkage drag where recombination is suppressed in the target gene region, and standard recombination-based approaches cannot be used in the molecular dissection of the target genes. In addition to this isolation of desirable genes from species belonging to secondary and tertiary gene pools is still difficult through map-based cloning. Although the wild species are good genetic resources for genes controlling resistance to biotic and abiotic stresses as well as quality traits, these could not be used extensively in breeding programs in either way.

Expression of Alien Genes: HGT through genetic transformation is one of the most exciting approaches that opened practical opportunities for the improvement of crop plant without any limitation of genome boundaries. However, the unpredictable silencing or variable expression of transgenes is a ubiquitous phenomenon and it is an important challenge for genetic engineering of crops. This has been observed invariably in all plant species studied. There is not yet a reliable way to prevent silencing, although the converse affects consistent gene silencing has been reported. Silencing resulting from interactions among multiple copies of transgenes and related endogenous genes involves homology based mechanisms that act at either the transcriptional or post transcriptional level. It has been shown that high level expression of foreign proteins in plants often leads to gene silencing.

Gene Flow: Flow of alien transgenes from transgenic plants to their weedy and wild relatives through sexual reproduction and/or vegetative propagation is one of major concerns with potential ecological risks. Gene flow from the transgenic plants having resistance to diseases and insect-pests, drought and salt tolerance and herbicide resistance can significantly enhance the ecological fitness of weedy and wild populations. As results, they can become aggressive weeds that can have unpredictable consequences to local ecosystems. This gene flow can also change the original wild populations and better ecological fitness could even lead to the extinction of endangered wild species populations locally Kiang *et al.*, (1992). Therefore, alien gene transfer through genetic transformation has a challenge of its negative impact of present ecological system.

New Genetic Approaches for Harnessing the Natural Variation: The domestication of the plant species for food, fodder or any commercial purpose for mankind is one of the very ancient practices. However, while carrying out domestication or breeding of any crop species, the gene pool has been narrowed with number of alleles (Tanksley and McCouch, 1997). Therefore, in general, breeders work with a limited number of alleles available in the cultivated gene pool and are unable to utilize the natural variation present in the germplasm collection of a particular species. In this context, wild species can serve as a reservoir of useful alleles to use them in breeding programme. Conventional methods of breeding, however, have limited scope as they render the transfer of only a fraction of the genetic variation from wild to cultivated species. Some selected approaches have been described below that have potential to utilize the alleles from wild species to breeding lines.

Introgression of Exotic Germplasm: Wild species together with landraces represent natural variation within the species. Domestication of these landraces which are highly heterogeneous in

nature is the first step to produce cultivars. Extensive studies have been done on the natural variation in crop species to study both evolutionary and ecological potential of the genes. It has been demonstrated that quantitative trait modification which includes phenotypic and compositional changes cannot be achieved by mutagenesis or transgenic but can be introgressed through wide genetic variation studies using molecular marker assisted breeding. Several other strategies have been used for introgression of favourable gene/QTL/chromosomal segment by developing isogenic lines using wild species and the variety /genotype of interest. Based on the protocol used for the development, the generated lines are referred as introgression lines (ILs), back-cross recombinant inbred lines (BCRIL) recombinant chromosome substitution lines (RCSLs) chromosome segment substitution lines (CSSLs). Introgression/ exotic libraries are constructed using introgression lines each of which carries a fragment of defined homozygous chromosomal segment from donor exotic parent with a homozygous genetic background of elite parent. These exotic libraries have been used for identification of QTLs controlling tomato aroma, fruit nutrition and antioxidant content. In rice, a large set of CSSL libraries were constructed which resulted in the transfer of brown plant hopper (BPH) and the white-backed plant-hopper (WBPH) resistance in the line. Hence, this approach can be employed to enrich the genetic variation which was lost during the domestication of crop plant.

Advanced-backcross (AB-QTL) Analysis: AB-QTL analysis is an approach for simultaneous discovery and transfer of QTLs from a wild species to a crop variety which was proposed earlier by Tanksley and Nelson. In this approach, a wild species is backcrossed to a superior cultivar, and during backcrosses, the transfer of desirable gene/QTL is monitored by employing molecular markers. The segregating BC2F2 or BC2F3 population is then used not only for recording data on the trait of interest, but also for genotyping using polymorphic molecular markers. These data are then used for QTL analysis, leading to simultaneous discovery of QTLs, while transferring these QTLs by conventional backcrossing. Many AB- QTL studies concluded that wild species contain favourable alleles for enhancement of quantitative traits for cereal crops.

Association Genetics: Association genetics is an approach that utilizes natural variation and linkage disequilibrium (LD) existed in natural population to identify the gene(s) / genomic regions associated with trait. In general, conventional linkage analysis using a bi parental mapping population such as F2 lines, back cross (BC) population and recombinant inbred lines (RILs) is a commonly used method for trait mapping. However, such mapping populations are derived from a few cycles of recombination events, hence limit their resolution of genetic maps and localize QTLs from 10 to 20 cm intervals and also do not essentially use the germplasm that is being actively used in breeding programs. In contrast, association mapping, based on LD measures the degree of non-random association between alleles at different loci. It does not require a segregating population and in some cases more powerful than linkage analysis for identifying the genes responsible for the variation in a quantitative trait. Association mapping offers three advantages over traditional linkage analysis– (i) increased mapping resolution, (ii) greater allele number, and (iii) reduced research time. Mapping based on LD allows for large scale assessment of allele/trait relationship when combined with a correction for population

structure. Under this approach, association between marker and trait is only expected when a QTL is tightly linked to the marker because the accumulated recombination events occurring during the development of the lines will prevent the detection of any marker/trait association in any situation where the QTL is not tightly linked to a molecular marker. Based on the scale and focus of a particular study, association mapping employs one of following two approaches:

- (i) Candidate-gene association mapping, which relates polymorphisms in selected candidate genes that have purported roles in controlling phenotypic variation for specific traits.
- (ii) Genome-wide association mapping, or genome scan, which surveys genetic variation in the whole genome to find signals of association for various complex traits.

Although candidate gene sequencing across several hundreds of genotypes for selected genes using Sanger sequencing was an expensive task in past, use of pools of amplicons for a range of genotypes and/or genes through NGS technologies is expected to reduce the costs significantly.

Multi-Parent Advanced Generation Inter-Cross (MAGIC) population: MAGIC population is a second generation mapping resources for crop improvement which is constructed using multiple parents. This mapping population strategy involves linkage and association methodology for mapping genetic variation in population segregating for multiple QTLs. The MAGIC which is constructed using multiple parents. This mapping population strategy involves linkage and association methodology for mapping genetic variation in population segregating for multiple QTLs. The MAGIC approach can be considered superior to association and linkage mapping as it allows both coarse and fine mapping of RILs developed from multiple parents by sampling seed of any generation with greater genetic variation. It has been demonstrated that if a large set of RILs are produced, the complex architecture of many traits which are associated with crop yield and quality can be studied using epistatic interactions. Furthermore, the MAGIC lines may show extensive segregation for plant developmental traits like plant height, maturity that may limit the use of MAGIC population in dissection of complex traits.

Next Generation Sequencing (NGS) Technologies: Detection and utilization of genetic variation has been a major task for plant breeders. Though, classical molecular markers such as RFLPs (restriction fragment length polymorphisms), RAPDs (random amplified polymorphic DNA), AFLPs (amplified fragment length polymorphisms) and SSRs have been used extensively for this purpose, the SNP marker system that has capability to detect the variation at single base level, however have not been used in many crop species. One of the major limiting factor in this direction has been the higher costs involved in sequencing the genes/ transcriptomes/ part of genomes of related individuals for SNP discovery. Three major sequencing platforms that are currently being used in plant species include Genome sequencer FLX Roche/454 , Applied Biosystems SOLiD and Illumina Genome Analyzer.

These three platforms provide thousands of million sequence reads in a single run in reduced time and less costs as compared to conventional Sanger sequencing technology. Among these three approaches, FLX/454 platform is superior in terms of read length (about 400 bp) but is rather expensive in terms of cost when compared with the Solexa and AB SOLiD. Yet another

approach based on single molecule synthesis is gaining attention and is termed as 3rd generation sequencing. Apart from this many new sequencing technologies are emerging and/or are at their infant stages to facilitate genome wide marker discovery in both model/major and orphan crop species.

Sequence data generated for parental genotypes of the mapping populations by using NGS technologies can be used for mining the SNPs at large scale. While in case of model plant species or major crop species, it is easier to align the NGS data from individuals to the reference genome sequence data, if available or the transcript sequence data available through EST sequencing projects. In case of under-resourced crop species where appropriate or adequate sequence data are not available, the best possible strategy is to sequence the cDNAs with NGS technologies and then align with the transcript data of the species, if available or of the related major/ model crop species. In case, these SNPs have been derived from genes or genic regions, the corresponding markers are also referred as functional markers/FMs. Organic molecular markers/GMM. Apart from developing SNP markers, NGS technologies can be and are being used for other applications such as de novo sequencing, association mapping, alien introgression, transcriptome expression and polymorphism, population genetics, evolutionary biology and genome-wide assembly in several crop species. With the development of rapid and inexpensive sequence technologies, the efficiency and accuracy in sequencing have interpreted the genomic information of many plant species.

TILLING: To understand the function of gene(s) many approaches like RNAi, gene knockout, site \directed mutagenesis, transposon tagging etc. have been exploited for many years. All these techniques demand the use of transgenic material which is not always possible in many commercially important crops. So it is not only impeding the functional analysis of gene(s) but also retards the improvement of existing as well as the development of improved cultivars. In this review, a non-transgenic technique called Targeting Induced Local Lesions IN Genomes (TILLING) was emphasized that determines the allelic sequence of induced point mutations in gene (s) of concern. It allocates the rapid and cost- effective detection of induced point mutations in populations of physical/chemically mutagenized individuals. Ethyl methane sulfonate (EMS) is mostly used and produce G/C to A/T transition by alkylating the G residues and the alkylated G resides to base pair with T instead of pairing with C.

Tilling (Targeting induced local lesions in genomes) a newly developed general reverse genetic strategy helps to locate an allelic series of induced point mutations in genes of interest. It allows the rapid and inexpensive detection of induced point mutations in populations of physically/chemically mutagenized individuals. In addition to allowing efficient detection of mutations by TILLING approach, EcoTILLING technology is also ideal for examining natural variation. Endonuclease CEL I cut effectively the multiple mismatches in a DNA duplex. Therefore, heteroduplex DNA of unknown sequence with to that of a known sequence reveals the positions of polymorphic sites. Both nucleotide changes and small insertions/deletions are identified. It can be performed more inexpensive than full sequencing the methods currently used for the most single nucleotide polymorphism (SNP) discovery. SNP variation can provide clues

to the adaptive strategies and population history that undoubtedly played roles in species evolution. It is also used for screening and detection of plants with desired traits by knockdown and knockout mutations in specific genes. This makes TILLING and EcoTILLING an attractive strategy for a wide range of applications from the basic functional genomic study to practical crop breeding.

TILLING Antiquity: It was first explored in the late 1990's by the efforts of Claire McCallum and his collaborators (Fred Hutchinson Cancer Research Center and Howard Hughes Medical Institute) who was experimenting on *Arabidopsis*. He used T-DNA lines and antisense RNA as reverse genetic approaches to illustrate the function of two chromo methylase genes. He was impotent to successfully apply these methodologies to describe the function of CMT2 gene. This technique was developed by pooling chemically mutagenized plants together, creating hetero duplexes among the pooled DNA, intensify the region of concern and using dHPLC (denaturing high performance liquid chromatography) to identify the mutants by chromatographic variations. A less expensive and faster modification of the TILLING protocol was published later, which uses a mismatch specific celery nuclease (CEL I) and LI-COR gel analyzer system. In 2001, the standard procedure was developed with practical software that makes the TILLING technique a more routine method to detect mutations to get satisfactory results.

The application of TILLING to crop improvement may also help with another constraint in domesticated species genomes having limited genetic variation. During domestication and subsequent selection, much of the genetic variation available in the wild crop progenitors has been lost. Thus, plant breeders have at times used wild relatives or land races to introduce useful genetic variation. This practice has been successful in wheat for developing disease resistant and higher yielding varieties and a land race was also used for the development of the first full waxy line because it carried a rare deletion allele of one of the waxy loci (Gilchrist and Haughn, 2005).

Eco TILLING: An extension of TILLING is EcoTILLING, which uncover natural alleles at a locus contrary to induced mutations. Many crop species cannot be exposed to induced mutation, and Eco TILLING can be used to find the natural variants and their putative gene functions in these crops. This can be done at low cost of SNP/haplotyping methods and one can screen many samples with a gene of interest. It does not require all the population to be screened to find the polymorphism which can be arduous and time consuming in TILLING. Moreover, in open pollinated crops, Eco TILLING can be used to find the heterozygosity level within a gene fragment. As the CEL I can digest small proportion of the hetroduplexes, it can be used to find the multiple polymorphisms in a single gene of interest. Furthermore, phylogenetic diversity estimates, selection and linkage disequilibrium can be estimated. It can be used to detect the DNA polymorphism in satellite repeat numbers. It can also be effectively used as an efficient, rapid technique to identify DNA polymorphisms in populations with high genetic similarity and to mine for SNPs in collections of plant germplasm. The EcoTILLING approach first time used on *Arabidopsis thaliana* in 2004 to mine for natural polymorphisms. EcoTILLING in *Mla* resistance genes of *Hordeum vulgare* (barley) was used in 2006 to examine the allelic variation. It demonstrates how effectively it can be used for the evaluation of diversity in natural

populations. It was employed to identify polymorphisms in mung bean (*Vignaradiata* (L.)). The majority of the polymorphisms were detected between *sublobata* and *radiata* in putative introns. In general, EcoTILLING shows great promise in the process of identifying natural disease resistance alleles, which can be used in crop improvement. Besides, this technique is applicable to any organism even those that are heterozygous and polyploidy in nature.

Role of Molecular Markers in Alien Gene Introgression

(1). Marker-Assisted Selection and Marker Assisted Backcrossing: Once the markers associated with a trait of interest is identified through linkage mapping, association mapping, AB-QTL or transcriptomics approach, etc., the next step is to use these markers in the breeding programme. In this context, the selection of one or a few genes (QTLs) through molecular markers using backcrossing is a highly efficient technique. There are three levels of MABC or MPS:

- (i) Foreground selection: which includes screening of target gene or QTL using molecular markers, this step can also be used for selection of recessive allele for backcrossing as recessive alleles require one generation of selfing for its expression.
- (ii) Recombinant selection: involves selection of the BC progeny containing the target gene and recombination events (between the target locus and linked flanking markers. The purpose of this selection step is to minimize the linkage drag by using markers that flank the target gene. This linkage drag poses a big problem during selection through conventional breeding methods. Furthermore this recombination selection event is usually carried out using two BC generations.
- (iii) Background selection: involves use of markers that are unlinked to the target locus for the selection of BC progeny containing highest proportion of recurrent parent (RP). In summary, the MABC employs linked markers to select the target gene/QTL from the donor parent and the unlinked markers to recover RP. Traditional approaches of recovery of RP genome take upto six BC generations but the use of markers enables to achieve the same in even in BC2.

(2) Marker-Assisted Recurrent Selection: There are cases where quantitative variation is controlled by many genes (QTLs) with minor effect; in such cases the previous approach (MABC) has certain limitations in the introgression of the target trait. Moreover, the markers identified linked with a trait to be used in MABC are generally identified biparental mapping populations. This limits the study of allelic diversity and genetic background which are very essential in crop breeding program. Limited statistical tools for studying polygenic traits controlled by many small effect loci are yet another drawback of MAS.

Furthermore, minor QTLs show an inconsistent QTL effect. Even though the effect of these minor QTLs is consistent, introgression of these QTLs through MABC approach becomes extremely difficult as a larger number (sometimes unmanageable) of progenies, depending on the number of QTLs, are required to select appropriate lines in MABC. In cases as mentioned, marker assisted recurrent selection (MARS) can be used for pyramiding of several genes/QTLs (of minor effect) in a single genotype. MARS is based on ad hoc significance test which include

the identification of trait associated markers and estimation of their effect. The approach involves multiple cycles of marker based selection that includes, (i) Identification of F₂ progeny which contain favourable alleles for most if not all QTLs, (ii) Recombination of the selected progenies to the selfed ones and (iii) Repetition of these cycles.

According to the recent studies, the response of MARS is larger in case of prior knowledge of the QTLs and the response decreases as the knowledge of the number of minor QTL associated with the trait decreases. In sweet corn, MARS was employed to fix six marker loci in two different F₂ populations which showed an increase in the frequency of marker allele from 0.50 to 0.80. Similarly in a separate study, enrichment of rust resistance gene (Lr34/Yr18) with the increase in frequency from 0.25 to 0.60 was reported in wheat BC1 through MARS. MARS, becoming a popular approach, can thus be effectively utilized for selection of traits associated with multiple QTLs by increasing the frequency of favourable QTLs or marker alleles.

(3) Genome-Wide Selection: Genome-wide selection is another approach that can be used to pyramid favorable alleles for minor effect QTLs at whole genome level. Unlike MABC or MARS, the GWS calculates the marker effects across the entire genome that explains entire phenotypic variation. The genome wide marker data (marker loci or haplotypes) available or generated on the progeny lines, therefore, are used to calculate genomic estimated breeding values (GEBV). It is important to note that the GEBVs are calculated for individuals based on genotyping data using a model that was „trained“ from individuals having both phenotyping and genotyping data. These GEBVs are then used to select the progeny lines for advancement in the breeding cycle. In summary, the GWS provides a strategy for selection of an individual without phenotypic data by using a model to predict the individual breeding value.

Conclusion:

Recent advances in sequencing and genotyping technologies have made it possible to develop molecular markers as well as undertake genotyping at large scale in both major as well as minor (or so called orphan crop species) that can be used not only for developing high density genetic and physical maps but also for generating transcriptome or sequence data. These approaches together with approaches such as transcriptomics, genetical genomics, metabolomics and proteomics can be used to identify the genomic regions or genes involved in expression of trait(s) that are of interest to the breeding community. In parallel, the high throughput sequencing and genotyping approaches can be used to detect genetic variation existed in germplasm collection not only in cultivated gene pool but also in landraces and wild species. TILLING and EcoTILLING are inexpensive and swift natural polymorphism detection and genotyping methods. They have advantages for determining the range of variation for genetic mapping based on linkage analysis. Such kind of genetic variation (or favourable alleles) can be introgressed in elite variety or genotype of interest by using AB-QTL approach or developing introgression libraries. Furthermore, the QTLs or genes or superior alleles for the trait of interest identified through linkage mapping, association mapping, AB -QTL approach or -omics approach can be

introgressed or pyramided in elite varieties or genotype of interest by using MAGIC, MABC, MARS or GWS approaches.

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INDIA AS A LEADER IN FARMING AMID THE GLOBAL FARMERS CRISIS

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Introduction:

In the 21st century, the agricultural sector faces unprecedented challenges globally, from climate change and soil degradation to water scarcity and economic pressures. Amidst these challenges, India has emerged as a crucial player in global agriculture, leveraging its historical agricultural heritage, technological advancements, and policy innovations. This chapter explores how India is positioning itself as a leader in farming, addressing global agricultural crises, and providing a model for sustainable agricultural practices. As the global agricultural sector grapples with unprecedented challenges ranging from climate change to economic volatility, India has emerged as a beacon of resilience and innovation. With its rich agricultural history, diverse agro-climatic zones, and a large population dependent on farming, India's approach to tackling the contemporary farmers' crisis offers valuable insights and solutions that can be emulated worldwide.

As global concerns about food security and sustainable farming intensify, India's experiences and strategies offer valuable lessons. The country's approach to managing water resources, promoting organic farming, and ensuring farmer welfare through social safety nets can serve as models for other nations. By fostering a collaborative environment that encourages knowledge exchange and technological transfer, India can lead the way in forging a more sustainable and equitable global agricultural system.

The Global Farmers Crisis

Globally, farmers are grappling with a myriad of issues that threaten food security and agricultural sustainability. These are includes here:

1. **Climate Change**

Erratic weather patterns, increased frequency of extreme weather events, and shifting growing seasons are adversely impacting crop yields.

2. **Soil Degradation**

Intensive farming practices, deforestation, and pollution have led to significant soil degradation, reducing agricultural productivity.

3. **Water Scarcity**

Over-extraction of groundwater and inefficient irrigation practices have resulted in water shortages, particularly in arid and semi-arid regions.

4. **Economic Pressures**

Farmers worldwide are struggling with low crop prices, high input costs, and mounting debt, exacerbating rural poverty and leading to high rates of farmer suicides (Rawat *et al.*, 2022).

5. **Technological Lag**

Many farmers, especially in developing countries, lack access to modern agricultural technologies, hindering their ability to improve productivity and resilience.

India's Agricultural Landscape

India, with its vast and diverse agricultural sector, is uniquely positioned to address these global challenges. Agriculture is the backbone of the Indian economy, employing over 50% of the workforce and contributing significantly to Gross Domestic Product (GDP). India's agricultural sector is characterized by (FAO, 2021):

1. Diverse Climates and Soils

India's varied climates and soil types allow for the cultivation of a wide range of crops, from staples like rice and wheat to cash crops like cotton and sugarcane.

2. Smallholder Farmers

The majority of Indian farmers are smallholders, with less than two hectares of land, presenting unique challenges and opportunities in agricultural development.

3. Rich Agricultural Heritage

India has a long history of traditional farming practices and crop varieties, which provide a foundation for sustainable agriculture.

Technological Advancements and Innovations

India is harnessing technology to revolutionize its agricultural sector. Key advancements include (International Water Management Institute (IWMI), 2022):

1. Precision Farming

The use of satellite imagery, drones, and global positioning system (GPS) technology is helping farmers optimize resource use, improve crop yields, and reduce environmental impact. Mobile applications and digital platforms are providing farmers with real-time weather forecasts, market prices, and expert advice, empowering them to make informed decisions.

2. Biotechnology

Advances in biotechnology, such as genetically modified crops and bio-fertilizers, are enhancing crop resilience to pests, diseases, and climate stress (Giller *et al.*, 2021).

Policy Initiatives

The Indian government has implemented several policy measures to support farmers and promote sustainable agriculture (Ministry of Agriculture & Farmers Welfare, Government of India, 2023)

1. Pradhan Mantri Fasal Bima Yojana (PMFBY)

This crop insurance scheme provides financial protection to farmers against crop losses due to natural calamities, pests, and diseases.

2. Soil Health Card Scheme

This initiative aims to promote soil health by providing farmers with soil health cards that contain information on nutrient status and recommendations for appropriate soil amendments.

3. Pradhan Mantri Krishi Sinchai Yojana (PMKSY)

This scheme focuses on improving irrigation infrastructure and promoting efficient water use practices through micro-irrigation systems.

Sustainable Agricultural Practices

India is leading the way in promoting sustainable agricultural practices that address global environmental concerns:

1. **Organic Farming**

India has become one of the largest producers of organic food, with initiatives to promote organic farming through subsidies, certification, and market linkages.

2. **Agroforestry**

Integrating trees and shrubs into agricultural landscapes is helping to enhance biodiversity, improve soil health, and sequester carbon.

3. **Integrated Pest Management (IPM)**

IPM practices are reducing the reliance on chemical pesticides, promoting the use of biological control agents, and improving crop health (Pingali *et al.*, 2019; Das *et al.*, 2024).

Case Studies

1. **Zero Budget Natural Farming (ZBNF):** Originating in Andhra Pradesh, ZBNF is a sustainable farming model that reduces input costs by using natural farming techniques. It has gained popularity among smallholder farmers and is being promoted nationwide. (Raj, 2019)

2. **The Green Revolution:** The Green Revolution of the 1960s and 1970s transformed India from a food-deficient country to a self-sufficient one. The adoption of high yielding variety seeds, fertilizers, and irrigation significantly boosted agricultural productivity (Pingali, 2012; Sunil *et al.*, 2023).

Challenges and Future Directions

Despite its progress, India's agricultural sector faces ongoing challenges (Singh and Sharma 2020):

1. **Climate Vulnerability**

Indian agriculture remains highly vulnerable to climate change, necessitating continued investment in climate-resilient practices and technologies.

2. **Economic Viability**

Ensuring the economic viability of smallholder farming through better market access, fair pricing, and financial inclusion remains a priority.

3. **Resource Management**

Sustainable management of natural resources, particularly water and soil, is critical to the long-term sustainability of agriculture.

Future directions for India's leadership in agriculture include:

1. **Strengthening Research and Development:** Investing in agricultural research to develop new technologies, crop varieties, and farming practices that address emerging challenges.

2. **Promoting Farmer Cooperatives:** Encouraging the formation of farmer cooperatives to enhance bargaining power, reduce costs, and improve access to markets and resources.

3. **International Collaboration:** Sharing India's agricultural innovations and experiences with other developing countries to contribute to global food security and agricultural sustainability.

Conclusion:

India's role as a leader in global agriculture is increasingly recognized, particularly as the world faces complex and interconnected challenges in the farming sector. Through technological innovation, sustainable practices, and supportive policies, India is not only addressing its own

agricultural challenges but also providing a model for other nations. As the global community seeks solutions to ensure food security and environmental sustainability, India's experiences and strategies offer valuable lessons and hope for a more resilient and sustainable agricultural future. (Shiva, 2004; Dubey *et al.*, 2021).

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SOIL UNDER SIEGE: UNDERSTANDING AND MITIGATING CLIMATE CHANGE EFFECTS ON SOIL HEALTH

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Abstract:

The profound impact of climate change, driven by global warming and anthropogenic activities, on soil properties is critical for agricultural productivity, environmental benefits, and overall soil health. This study synthesizes the latest research on how climate change affects soil temperature, moisture, organic carbon content, pH, structure, and biodiversity. Elevated soil temperatures accelerate organic matter decomposition, affecting nutrient cycling and carbon stocks. Changes in precipitation and evapotranspiration alter soil moisture, leading to deficits that hinder microbial activity and nutrient availability. Variations in temperature and precipitation patterns also influence soil pH, structure, and erosion, disrupting soil health and fertility. Soil biodiversity, crucial for ecosystem functioning, faces significant changes due to altered habitat conditions and food availability. This integrative approach highlights the interconnectedness of soil properties and the need for interdisciplinary strategies to address climate-induced soil challenges. Enhanced scientific understanding from this synthesis can inform policymakers and soil scientists, improving agricultural practices and environmental benefits preservation. The chapter emphasizes the necessity of adaptive management practices and policies to enhance soil resilience and sustainability, thereby contributing to climate change mitigation efforts.

Keywords: Climate Change, Soil Health, Agricultural Productivity, Environmental Benefits, Soil Biodiversity, Interdisciplinary Strategies.

Introduction:

Climate change, driven by global warming and anthropogenic activities, profoundly impacts various environmental processes, including soil properties, which are critical for agricultural productivity, ecosystem services, and overall soil health (IPCC, 2014). This chapter provides a comprehensive synthesis of the latest research on the impacts of climate change on soil properties, emphasizing their multidimensional nature and broader implications. Soil properties such as temperature, moisture, organic carbon content, pH, structure, and biodiversity are all susceptible to climatic changes, and alterations in one can lead to cascading effects on others (Lal, 2004). The novelty of this study lies in its integrative approach, highlighting the interconnectedness of soil properties and the need for interdisciplinary strategies to address climate-induced soil challenges. Enhanced scientific understanding from this synthesis can inform policymakers and Soil Scientists, improving agricultural practices and ecosystem services preservation, while contributing to climate change mitigation efforts (Schmidt *et al.*, 2011;

Banwart, 2011). By synthesizing current research, this chapter advances our understanding of climate change effects on soil properties and suggests directions for future research, aiming to inform adaptive management practices and policies that enhance soil resilience and sustainability (Nearing *et al.*, 2004; Lal, 2010).

1. Soil Temperature

Climate change significantly impacts soil temperature regimes by increasing atmospheric temperatures and altering precipitation patterns. Elevated soil temperatures accelerate microbial activity, leading to faster organic matter decomposition and nutrient cycling. This can reduce soil organic carbon (SOC) stocks, negatively affecting soil structure and fertility. Schlesinger *et al.*, (2009) demonstrated that higher soil temperatures speed up organic matter decomposition, potentially releasing significant amounts of CO₂ into the atmosphere.

The combination of higher air temperatures and reduced snow depth has enhanced spring soil temperature trends, while winter soil temperatures show no significant trends. A study by Zhan *et al.*, (2005) found that soil temperature responses in Canada during the twentieth century varied by -2° to 7°C from atmospheric temperatures due to influences such as snow cover, precipitation, and vegetation. This variability highlights the necessity for integrated models that combine atmospheric, snow, soil, and ecological dynamics to accurately predict the impacts of climate change.

Since the late 1950s, significant soil warming in Canada has been primarily driven by spring and summer warming, with winter soil warming often undetectable due to reduced snow cover. The reduction in snow cover affects soil insulation, influencing soil temperature trends and underscoring the importance of considering snow insulation in climate change projections (Qian *et al.*, 2011).

2. Soil Moisture

Changes in precipitation patterns, coupled with increased evapotranspiration rates due to higher temperatures, significantly impact soil moisture levels. In regions experiencing reduced rainfall or prolonged droughts, soil moisture deficits can lead to several adverse effects, including reduced microbial activity, hindered nutrient availability, and increased soil compaction and erosion. Trenberth *et al.*, (2014) highlight that climate change-induced alterations in precipitation patterns are leading to more frequent and intense droughts, which significantly affect soil moisture and agricultural productivity.

Recent studies have advanced our understanding of how soil moisture influences climate. Building on initial research from the 70s and 80s, these studies use multiple models to improve the accuracy of predictions and assessments (Seneviratne *et al.*, 2010). According to Grillakis (2019), climate change is projected to increase the frequency of extreme and severe soil moisture drought events, further emphasizing the importance of understanding soil moisture dynamics in the context of climate change.

3. Soil Organic Carbon (SOC)

Soil organic carbon (SOC) is crucial for soil health, contributing to nutrient availability, soil structure, and water retention. Figure 1 depicts that climate change affects SOC dynamics by altering plant biomass production, litter decomposition rates, and soil respiration. As temperatures rise and precipitation patterns change, SOC decomposition increases, leading to reduced carbon sequestration in soils, which undermines efforts to mitigate climate change (Lal, 2004).

While land use change plays a significant role in SOC changes, climate change also contributes, albeit differently across regions (Zhou *et al.*, 2019). Studies indicate that climate change, particularly increasing temperatures and decreasing precipitation, influences SOC levels, although land use and management practices have a greater impact (Fantappiè *et al.*, 2011). Recent research highlights that climate and vegetation affect 20–40% of SOC turnover, with faster turnover in low latitudes and slower turnover in high latitudes. Additionally, long-term climate conditions affect passive SOC pools, reflecting changes in soil maturity and mineralogy (Trumbore, 1997).

Changes in disturbance frequency due to climate change can significantly affect the balance of carbon storage and release in terrestrial ecosystems, emphasizing the complex interactions between climate, land use, and SOC dynamics (Susan, E.T, 1997).

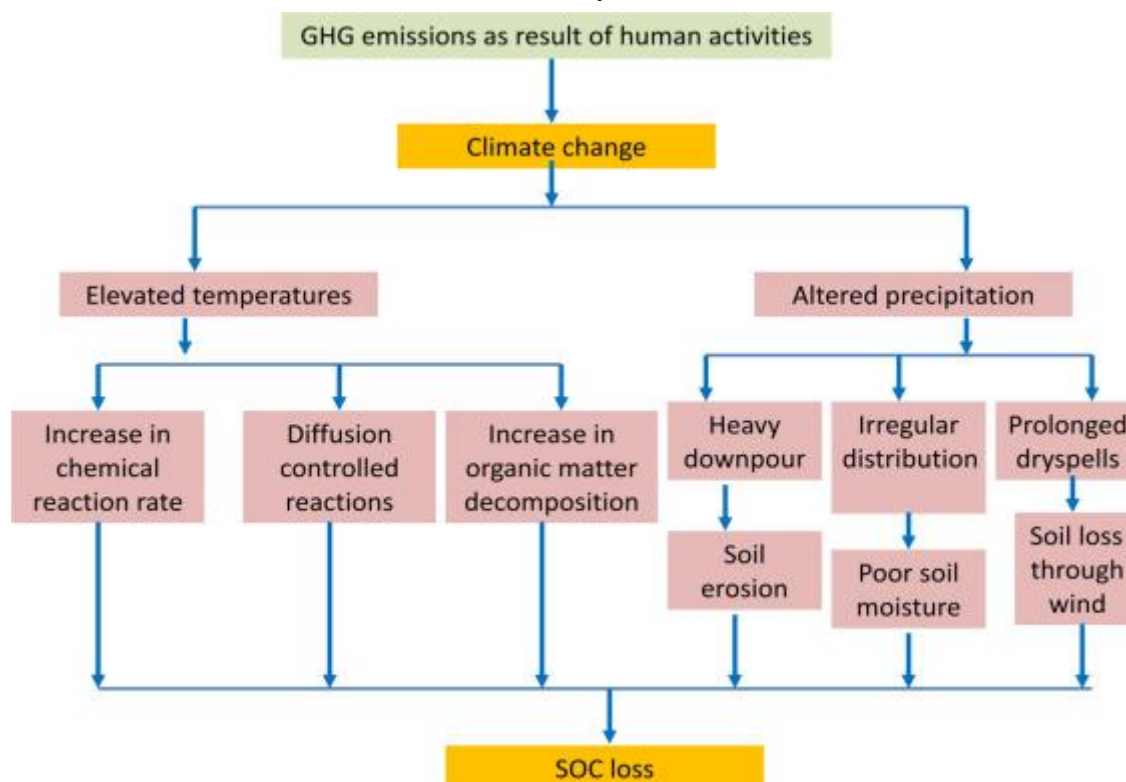


Figure 1: Soil organic carbon (SOC) as affected by Climate change
(Source: V. Girija Veni *et al.*, 2020)

4. Soil pH

Changes in precipitation patterns and increased atmospheric CO₂ levels can influence soil pH. Acid rain, resulting from higher concentrations of atmospheric pollutants, can lower soil pH, affecting nutrient availability and microbial activity. Conversely, reduced rainfall in arid regions can lead to soil alkalinity. For instance, the study by Johnson *et al.*, (1991) found that acid rain significantly lowers soil pH, leading to nutrient imbalances and reduced forest productivity.

Variable and changing climate can impact soil properties, especially pH, which affects all other ecosystem properties. Specific effects of altered temperature and rainfall patterns on soil properties vary depending on initial soil and ecosystem characteristics. Increased biomass production in managed ecosystems due to higher temperatures or rainfall may lead to soil acidification through the removal of alkalinity in harvested produce and increased leaching of basic cations, as highlighted by Rengel Z. (2011). Understanding these effects is crucial to prevent the loss of environmental function in soils and ecosystems.

5. Soil Structure and Erosion

Climate change has a significant effect on soil structure and erosion, primarily due to the increased intensity and frequency of extreme weather events, such as heavy rainfall and storms. These events disrupt soil aggregates and degrade soil structure, reducing its capacity to retain water and nutrients (Figure 2). Varallyay, G. Y. (1990) illustrated how climate change influences soil moisture, texture, structure, and erosion, driven by changes in temperature, precipitation, and vegetation patterns.

Studies by Nearing *et al.*, (2004) have highlighted that climate change-induced increases in rainfall intensity can substantially raise soil erosion rates, posing a threat to soil health and agricultural sustainability. With global warming expected to increase total rainfall and the frequency of intense rainfall events, soil erosion rates are projected to be significantly impacted. Changes in rainfall patterns, along with temperature, solar radiation, and atmospheric CO₂ concentrations, affect erosion processes, including rainfall erosivity, runoff, and soil susceptibility to erosion.

Recent research suggests that despite decreased annual rainfall, rainfall erosivity levels may increase, leading to higher erosion and runoff rates. Additionally, farmers' responses to climate change could either exacerbate or mitigate these effects. Soil erosion, influenced by changing rainfall patterns, results in the depletion of soil organic carbon (SOC), impacting soil fertility and carbon sequestration. Studies like Mondal A. *et al.*, (2016) quantify the future impact of climate change on soil erosion and SOC, indicating increased erosion and SOC loss rates in areas with steeper slopes, sandy soil, and fallow lands.

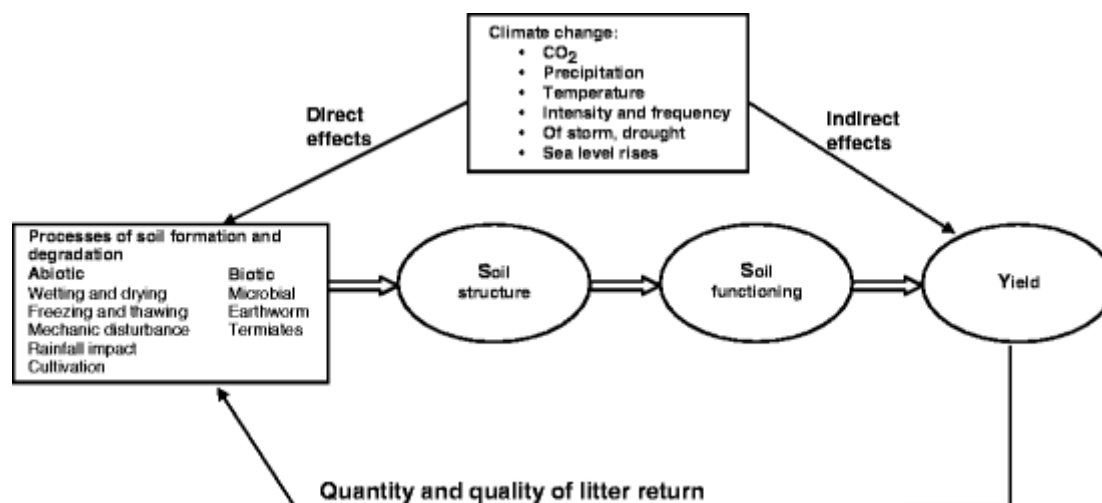


Figure 2: Effect of climate change on soil structure and soil health
(Source: Chan K.Y, 2011)

6. Soil Biodiversity

Soil biodiversity, encompassing microorganisms, fungi, and fauna, plays a vital role in preserving soil health and ecosystem functioning. Climate change can disrupt soil biodiversity by altering habitat conditions, food availability, and interactions among soil organisms. Changes in soil moisture, temperature, and organic matter inputs can lead to shifts in microbial community composition and activity.

Research by Mandal and Neenu (2012) demonstrated that global climate change significantly affects soil biodiversity and related services by modifying climatic parameters like temperature and humidity, thereby impacting various microorganisms and microfauna in soils. With soil biodiversity comprising diverse organisms such as fungi, bacteria, algae, and viruses, changing disturbance regimes under climate change pose challenges in assessing and predicting soil community responses.

Bardgett and van der Putten (2014) discussed the influence of climate change on soil microbial communities, highlighting implications for ecosystem services and soil health. Climate change also alters plant-soil feedbacks (PSFs), influencing plant performance, diversity, and ecosystem processes through changes in species distributions and community composition. These interactions, influenced by temperature, moisture, and other factors, exhibit context-dependent effects on vegetation-soil-climate feedbacks (Pugnaire F.I. *et al.*, 2019).

Understanding the impact of climate change on soil microbiomes and their ecosystem services is a critical challenge and opportunity in addressing pressing environmental problems, as explored in the review by Jansson and Hafmockel (2020) across various climate-sensitive soil ecosystems.

Conclusion:

Addressing the multifaceted challenges posed by climate change on soil health necessitates integrated approaches and adaptive strategies. The impacts of climate change on soil

properties, including temperature, moisture, organic carbon, pH, structure, and biodiversity, have far-reaching implications for agricultural productivity and ecosystem services.

By synthesizing current research findings, this chapter highlights the importance of interdisciplinary collaboration and scientific research in understanding and mitigating these impacts. Sustainable land management practices and adaptive strategies are essential to enhance soil resilience and mitigate adverse effects on agricultural systems and natural ecosystems.

Furthermore, effective policymaking informed by scientific knowledge is crucial to support soil conservation efforts and promote sustainable land use practices. Investing in research and innovation to develop climate-resilient agricultural practices and soil management techniques is essential for maintaining soil health and productivity in the face of climate change.

Overall, addressing the challenges posed by climate change on soil health is imperative for ensuring food security, preserving ecosystem services, and promoting the long-term sustainability of agricultural and natural systems.

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DIFFERENT WAYS TO MANAGE CROP RESIDUE FOR SUSTAINING THE SOIL FERTILITY AND CROP PRODUCTION UNDER CLIMATE CHANGE

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Introduction:

The uneconomic portion of a plant (stem, leaves, straw, husk, roots, bagasse, and molasses) that remains in the field after the economical component (grain) of the field crops has been harvested and threshed is known as crop residue. One benefit of recycling crop residue is that it can be utilized to meet its fertilizer needs of next crop. Remaining crops residues feed plants and provide organic C to soil microbes. Getting together residue that is still visible on the surface of the soil can significantly lower soil erosion, runoff, soil evaporation, and land preparation expenses.

India is an agricultural country, so in addition to crops, a large amount of residue is produced. The country produces a substantial amount of crop residue each year i.e. 685 million tonnes (Bhattacharjya *et al.*, 2019). The Indo-Gangetic Plains (IGP) contain 90% of the land used for the Rice Wheat Cropping System (RWS), one of the most widely used cropping systems in India. Because combine harvesters have advanced, over **75%** of the rice fields in the northwest of the IGPs are harvested mechanically.

Crop residue management is the term for the environmentally sound and economically viable handling of crop residue. Most farmers remove the wheat straw in order to provide it to the animals. Rice straw is thought to be a poor animal feed due to its high silicon concentration, which also poses serious crop management issues. A vast swath of loose rice leftovers left by the combine harvester obstructs with the seed drill used for planting wheat. To avoid these problems, farmers burn the 90–140 Mt of agricultural waste they produce each year (Bhuvaneshwari *et al.*, 2019). Farmers claim that burning rice straw is the most effective method of getting rid of it. It is an excellent pest management strategy in addition to being a cost-effective method. Due to burning of residues plant nutrients and biomass, or organic carbon, are therefore significantly lost. There are also detrimental effects on the health of the soil's flora and fauna as well as its attributes.

Methods of Crop Residue Management

On the basis of the management site is mainly of 2 types:

- 1) On-farm management
- 2) Off-farm management

1) On-farm Management

This covers the management of residues in the field, or at the location where the residues are produced. Crop residues are the source of numerous major and minor plant nutrients that are necessary for plant growth. In India, the 685 million tonnes of crop residue have the potential to supply 5.5 million tonnes of NPK to the soil. Soil fertility and productivity can be increased through the appropriate management of crop residues.

a) Burning of Crop Residue:

Large amounts of these leftover agricultural wastes are burned on the fields, mostly to remove any leftover straw and stubbles following the harvest, which obstructs the tillage and sowing processes for the following crop. The primary causes of crop residue burning in the fields are an increase in the usage of combines for harvesting, a lack of labor, and the high cost of clearing waste from the field. Burning of crop residues causes the large loss of nutrients. In addition to getting rid of illnesses and pests, this process contaminates the air and significantly reduces the number of microorganisms in the soil. Burning wastes results in the loss of soil, organic substances, which are undoubtedly dangerous to the rice-wheat system's sustainability.

b) Residue Removal:

After the crop is harvested and threshed, the crop residue, or husk and straw, is removed from the field and put to numerous other purposes. These leftovers can be utilized as a raw material in a variety of sectors, as well as for animal food (such as wheat straw), fuel (such as cotton leftovers), and bedding for dairy and poultry. The removal of residues also eliminates organic carbon from the soil, which has a negative impact on soil microbial activity. It also throws off the cycle of nutrients. As thus, the residues are eliminated at a rate that is sustainable.

c) Surface Retention:

Surface retention is the process of leaving agricultural remains on the soil's surface after the crop has been harvested. The retention of residues on the soil's surface enhances its physical, chemical, and biological characteristics. Additionally, it lessens soil erosion and aids in the sequestration of carbon.

d) Mulching using Crop Residues:

In conservation agricultural practice the crop residues are chopped into smaller pieces and uniformly spread on the field. Retention of residue on the soil's surface seems to be a better option in terms of conserving soil and avoiding water loss through evaporation. Furthermore, it reduces the rate at which weed seeds germinate and facilitates the growth of soil microbial communities, which in turn raises soil organic carbon, a direct indicator of the health of the soil. In the rice-wheat system in the northwest Indo Gangatic Plain, zero-till wheat has improved profitability, resource efficiency, and wheat yield.

e) *In-situ* Residue Incorporation:

By using the various machineries the crop residues are incorporated into the soil which will add organic carbon to the soil ultimately improving the physical, chemical and biological

properties of soil. Activity of soil microbes will also be increased. Improves the water holding capacity of soil, increases infiltration, reduces the bulk density.

While crop wastes have the ability to recycle nutrients in the field, doing so can temporarily immobilize certain nutrients (nitrogen, for example), which means that additional nitrogenous fertilizer will need to be added. Additional N should be given in order to rectify the excessive C:N ratio that was present during residue integration. This brief nitrogen scarcity is brought on by decomposer microorganisms immobilizing available nitrogen from fertilizer and soil. This time depends on the quality of the residue, the soil environment, and how long it takes for crop residue to decompose before planting the following crop.

f) Composting:

Crop leftovers are heaped in dung pits and used as animal bedding to produce compost. In the animal sheds, every kilogram of straw absorbs roughly two to three kilograms of urine, which added N to make it better. One hectare of leftover rice crop debris can be composted to create about three tonnes of manure, which has the same nutritional value as farmyard manure (FYM). Utilizing low-grade rock phosphate that is readily available locally, the rice straw compost can be fortified with P to create value-added compost that contains 2.3% P₂O₅, 2.5% K₂O, and 1.5% N (Sindhu and Beri *et al.*, 2005).

2) Off-farm Residue Management:

Crop leftovers can be managed in a variety of ways, both off-farm and ex-situ. However, ex-situ operations are typically limited by high collecting costs and transport.

a) For Livestock:

Due to their high nutritional value, most crop wastes can be turned into animal feed. For example, wheat straw is extremely nutrient-dense, so it's gathered in large quantities, stored, and used as feed all year long. Any leftover straw is sold. Because of its low nutritional value and high (6–12%) silica content, rice straw is not a desirable choice for animal feed (Lohan *et al.*, 2018). Cattle's levels of calcium and phosphorus become unbalanced when they are fed rice straw.

b) For Mushroom Cultivation:

Growing mushrooms on leftover agricultural residue is a great way to turn it into food. Mushrooms contain an amino acid composition equivalent to that of milk or meat and two to three times the protein of other vegetables, despite their high moisture content. *Volvariella volvacea* (straw mushroom) and *Agaricus bisporus* (white button mushroom), two of the four most commonly produced fungus, grow nicely on wheat and rice straws. To cultivate *Agaricus*, which has a very high substrate conversion efficiency into fungal bodies, straw is typically combined with hay and horse manure.

c) For Biofuel Production:

Burning biofuel lowers reliance on fossil fuels without a doubt. Because ethanol can be added to gasoline to raise its octane and lengthen its fuel life, or used directly in internal combustion engines, the ligno-cellulosic biomass conversion process is crucial. It is estimated

that 382 to 471 t⁻¹ of dry matter are produced during the manufacturing of ethanol from a variety of feedstocks, including sawdust, bagasse, rice and wheat straw, and maize grain, depending on the source. However, India is advancing its method of producing ethanol from agricultural waste. For agricultural residues to be completely converted into alcohol, a few process processes need to be enhanced.

d) For Paper Production:

When making paper, 40:60 is the ratio of rice to wheat straw. Paddy waste is the sole optimal raw material used in the production of pulp and paper boards. The paper and pulp board sector uses leftover rice as a raw material in around half of its operations.

e) Biochar Production:

Biochar and crop wastes have drawn a lot of attention since as practical methods to maintain the health of the soil. Finely ground, high-carbon charcoal, or biochar, is produced by a process called pyrolysis, which involves gently heating biomass in the absence of oxygen at 300-600 degree celsius (Singh and Sindhu *et al.*, 2014). It may be critical to the soil's long-term ability to store carbon. Since biochar is made from plant biomass and includes a unique kind of carbon that is resistant to microbial destruction, it can be applied to soil to act as a carbon sequester. Additionally, research has shown that biochar can improve water quality by efficiently absorbing contaminants and reduce greenhouse gas emissions from agricultural regions. However, the pyrolysis conditions and the biomass sources often impact the biochar's characteristics.

Machineries used in Crop Residue Management:

1. Straw Chopper:

A straw chopper, which comes after a combine harvester, is used to make straw from the paddy stubbles. Paddy straw chopper is useful for repurposing unused straw in the field.

2. Straw Baler:

Compressing, handling, transporting, and storing cut and raked products including silage, hay, cotton, flax straw, and salt marsh hay are done with a baler, commonly known as a hay baler.

3. Super Straw Management System (SMS) Machine:

The super straw management technique combines crop harvesting and residue management into one operation by adding additional machinery to a combine harvester to cut standing stubble on the soil's surface into small pieces.

4. Happy Seeder:

The Happy Seeder is a tractor-mounted device that plants wheat in the ground, slices and lifts rice straw, and then spreads the mulch of straw over the planted area.

5. Superseeder:

Also known as the Ultimate One Pass Sowing Solution Machine, is a tractor-driven tool that simultaneously tills the soil, plants seeds, and eliminates paddy stubbles before combining the ingredients with soil.



Straw Chopper



Straw Baler



Happy Seeder



**Super Straw Management System (SMS)
Machine**



Superseeder

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CULTIVATION TECHNOLOGY OF DRAGON FRUIT

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1. Introduction:

Dragon Fruit, or Red Pitaya, is a very interesting cactus from Southern Mexico, Guatemala, and Costa Rica. It is a brilliant red, ranging from red to pink depending on the variety, making it look outstanding. Therefore, besides being an excellent ornamental plant, it is highly valued in terms of its content of food nutrients. In addition, it is economically valuable; hence, it is cultivated. Its many nutritional benefits, such as strong antioxidants, high levels of vitamin C, fiber content, and overall nutritional value, have earned it the label 'nutritionally nutritious food.' The dragon fruit can aid not only in the protection of the human immune system, but also in the potential treatment of diabetes. It can also be made into different products like juice, ice cream, and wine. It puts farmers at an advantage due to a prompt return, proving to be as short a period as within one year from planting because it demonstrates its production potential in three to five years. Dragon fruit is highly flexible in relation to climates and types of soils, provided there is proper drainage. This means that it can be a profit-yielding crop for farmers across many Indian states. Dragon fruit plantation is an upcoming profitable venture for low-income groups as well as an interest among farmers, and the cultivation of it is gaining popularity due to the exchange of knowledge and planting materials. As new dragon fruit farms keep causing success and creating stories of profit, it may be that this fruit contributes its bit to the decline in malnutrition by offering the farmers a source of new income. It could also assist in satisfying the increasing need for antioxidant-rich, functional fruits like the dragon fruit and might be significant for raising the profitability of local farmers. In general, the prospects of growing dragon fruit in India are high, and it might prove to be a beneficial strategy for crop diversification and exploiting market opportunities.

2. History and Origin:

Dragon Fruit, or Pitaya in its other taxonomy, welcomes its origin from Central and South America and thrives as a climbing epiphyte. In the course of time, the fruit has gained mammoth popularity worldwide, and in the tropical and subtropical zones, the cultivation of dragon fruit has spread to many countries. Growing the dragon fruit is relatively easy because it only needs strong drought resistance, the ability to adjust to varying light and temperature, and tolerance for varying levels of soil salinity. It has found on every tropical island; however, the name varies by region. It has called pitahaya in Mexico and in Central and Northern South America; meanwhile, it is known as dragon fruit in Mexico. Its market extends regionally; it has produced in more than 20 countries globally. Some include Indonesia, Colombia, Israel, the

Philippines, Malaysia, and southern China. The Dragon Fruit has come to have a certain reputation for coming from Vietnam. It was introduced to the country by the French a little more than a hundred years ago. Thanh Long may give this name its most profitable, but it is high-yielding around these parts, under the given nomenclature. The elliptical ones are red in the epicarp with green designs, like the skin of a dragon. Vietnam is currently the world's number-one largest exporter of dragon fruit, making a significant contribution to the country's total fruit export turnover. It is therefore still a key economic factor for Vietnamese farmers, as it is the most profitable crop.

Indian farmers' focus is slowly shifting toward dragon fruit. The plant is native to southern Mexico, Guatemala, and Costa Rica. It has first introduced in India during the late 1990s and slowly gained popularity in places where the climate is most suitable: dry and free from frost, mainly in southern, western, and north-eastern India. Farmers from many states of India have taken seriously growing Isabgol, which includes Karnataka, Kerala, Tamil Nadu, Maharashtra, Gujarat, Orissa, West Bengal, Andhra Pradesh, and the Andaman and Nicobar Islands. The above information is by and large only impressionistic, being gathered from field observations and press reports. On the other hand, the production zone, so far, has said to be quite small and reckoned to be less than 400 hectares all over. Much of the production space is in the form of small-scale cottage cultivation. The fruits that have imported mostly come from neighboring countries like Thailand, Malaysia, Vietnam, and Sri Lanka. Strong interest from the domestic and international markets points to a very promising future for cultivation.

3. Nutritional Composition:

People celebrate dragon fruit as a tropical superfood, packed with thousands of beneficial nutrients and low in calories. Being vied by much health consciousness, vitamin-rich and mineral-packed ripe dragon fruit is in demand for improved digestion, apart from its potential medicinal properties against cancer, diabetes, high cholesterol, and high blood pressure. Dragon fruit is a good source of important nutrients believed to be effective for managing chronic diseases, digestive health, and immunity maintenance. Nutrient-wise, it presents an ideal choice for weight loss, diabetes control, and hypolipidemia, carrying excellent nutritive profiles while providing various vitamins, minerals, and dietary fibers. There are two types of dragon fruit: white or red/pink flesh, with little calories but rich in phenolics, flavonoids, and antioxidants, which promote beneficial health for human consumption. Biochemical analysis revealed that dragon fruit has a moisture content of 83–88% and is slightly acidic, with a titrating acidity ranging from 0.20 to 0.30 mg lactic acid equivalents. The content of its soluble solids varies from 8 to 12 °Brix, and therefore the perceived sweetness by consumers also fluctuates. Moreover, dragon fruit is a rich source of vitamin C that ranges from 4 to 10 mg/100 g and contributes significantly toward the recommended daily amount. Also, pink-fleshed fruits contain more phenolics and flavonoids than white-fleshed fruits, which contributes more to antioxidant activity. Furthermore, other health-promoting phenolic acids highlight its functional food status, offering protective antioxidant effects and demonstrating the potential for health benefits. Besides, the high content of water and minimal supply of energy in the dragon fruit

have been elaborated by the USDA nutritional analysis, making it the right choice for a wholesome diet.

4. Botanical Description/ Species and Varieties:

Dragon fruit, belonging to family Cactaceae, characterized by climbing or epiphytic cacti with angular stems and mostly bland to bright white, fragrance, night-blooming flowers, is from Central and South America. The most common are *H. undatus* which is the one with red skin and white flesh, but also the commercialized type *H. polyrhizus* (red skin and red flesh) and *H. megalanthus* (yellow skin and with white flesh.) The genus *Hylocereus*- a genus that includes several species and varieties of Dragon Fruit each identified by different fruit qualities and growing characteristics as well. The first type has a red fruit coat and white pulp, while the second type has red fruit coat and red pulp, the third type is yellow fruit coat and white pulp and is not commonly used, the red leather one is more popular. Flowering in Dragon Fruit usually starts from small spiral button-like things at stem margins and ends up into flower buds within 10-15 days. The hermaphroditic flowers are 25 to 30 cm long with white inner wings, green-yellow outer calix colored by purple stains disc, scent violet and nocturnally flaring categories until dawn They flower in the main from May to September, and the picking season begins 30-40 days after the fruit has set.

Although fruit quality may differ between varieties, harvesting timing determines quality more than varietal differences. Both self-compatible and self-incompatible Dragon Fruit varieties can be found, being the majorities of the Asian varieties self-compatible. Various varieties sleeve fruit differently in terms of sizes (and some also in shape), while detailed information on varietal and production aspects are largely unavailable and still need to be defined. Main varieties grown - in the Philippines Which includes the one with white flesh, the *Hylocereus undatus* variety and what is a bit darker red fleshed, the *Hylocereus costaricensi* variety. Vietnam mainly produces white, red, and purple-pink dragon fruits. The purple-pink and red dragon fruits are a little bit sweeter compared to the white flesh variety. Also, the EU has great demand for Vietnamese dragon fruit.

4.1 Different kinds of Dragon fruits:

- 1. Sour Dragon Fruit** – The Sour Dragon Fruit, a member of the *Stenocereus* family, is native to arid locations throughout the Americas. The fruit has special appeal in the northern portion of Mexico, where residents persist in harvesting it. This fruit is renowned for its invigorating flavor that blends a piquant bite with a tart sourness.
- 2. White Dragon Fruit** – The White Dragon Fruit is the predominant kind of dragon fruit that is extensively grown. The fruit has a pink exterior and a succulent, white inside. Neitzel, a popular cultivar native to California, is particularly enjoyable when cold, providing a sweeter and more delectable taste.
- 3. Red Dragon Fruit** - This sweet variety of dragon fruit has both red skin and flesh. It has a texture like that of a kiwi. You can enjoy it raw or add it to your smoothies. Just be careful because eating red dragon fruit can leave stains on your hands.

4. **Yellow Dragon Fruit** - This type of dragon fruit has a yellow skin with a cool scaling pattern. The flesh is firm and white, and it is the tastiest variety, also known as Pitaya Amarillo. It's native to South America.
5. **Purple Pink Color Fruit** - This fruit has a pinkish color and its flesh is white. It is also known as Pithaya Blanca or white-fleshed pitaya.

4.2 Dragon Fruit Varieties

1. **Alice Dragon Fruit (*Hylocereus undatus*)** – This variety got its name from Alice Snow, a member of the rare fruit growing community in California. It's a medium-sized fruit, weighing around 0.5-1.0 lbs (0.23-0.45 kg), and has a semi-sweet taste. When fully ripe, the outer skin turns into a vibrant hot pink, making it one of the most visually striking varieties out there. However, to bear fruit, this variety requires cross-pollination.
2. **Guyute Dragon Fruit (*Hylocereus undatus*)** – Guyute is a fruit that falls in the medium to large size range, usually weighing between 0.75-1.0 lbs (0.34-0.45 kg). It boasts a delightful sweet flavor, perfect for enjoying when chilled and eaten fresh. The best part is, it doesn't need any external help for pollination—it can set fruit on its own.
3. **Halley's Comet Dragon Fruit (*Hylocereus undatus x polyrhizus*)** – Halley's Comet is one variety which has cross between white-fleshed and red-fleshed dragon fruit. It is large in size, generally weighing near 1.5-2.0 lbs (0.68-0.9 kg). The fruit is round in shape as compare to physical graffiti, and its shorter skin length. Other than these variations, they are largely identical in all other features.
4. **Harpua Dragon Fruit (*Hylocereus undatus*)** – Particularly these variety is a medium to large in size and weighs approximately one pound. It has wonderful milky white pulp which is sweet and incredibly refreshing. The flavor is like a mellow melon. And the best thing is it is self-pollinated variety.
5. **L.A. Woman Dragon Fruit (*Hylocereus undatus*)** – Now, this one's a wonderful looker, but not much more. It's got a smooth, mellow, and sweet flavor that'll certainly provide your taste buds a treat. However, it tends to low quality aftertaste. The fruit is medium to large in size, ranging from 0.75 - 1.5 lbs (about 0.34 to 0.68 kilogram).
6. **Lake Atitlan Dragon Fruit (*Hylocereus guatemalensis*)** – This variety is named after a lake in Guatemala that's surrounded by volcanoes in Central America. It's a medium to large fruit, usually weighing between 0.75 -1.0 lbs (about 0.34 to 0.45 kg). The pulp is packed with flavor – it's sweet and tangy at the same time. You can totally use this one as a garnish or mix it up with other juices. Plus, it can pollinate itself.
7. **Makisupa Dragon Fruit (*Hylocereus undatus x polyrhizus*)** – Makisupa is a fantastic fruit with a vibrant magenta flesh that has a sweet yet slightly tangy taste. It's a real eye-catcher, with its green fins spiraling around the pink surface. These fruits are medium to large, weighing up to one pound, and the best part is they can pollinate themselves. Originally from Thailand, this variety of dragon fruit made its way to Florida and is now grown on a small scale commercially in Homestead. It consistently produces delicious dark magenta fruits that usually weigh around one pound. The best part is, you don't need to worry about

cross or hand-pollination with this one, making it a great choice for both homeowners and commercial farms.

8. **Neitzel Dragon Fruit (*Hylocereus undatus*)** – Another gem brought to you by California's rare fruit growers, Neitzel is an incredibly appealing fruit with its white flesh and a sweet and irresistible flavor. These fruits are medium to large, weighing up to 1 lb (0.45 kg). They taste best when chilled and eaten fresh, but they also make a great garnish. And just like the Makisupa, this variety can take care of its own pollination.
9. **Physical Graffiti Dragon Fruit (*Hylocereus undatus* x *H. polyrhizus*)** – This variety is a hybrid of red-fleshed and white-fleshed fruit. The result is a remarkably delicious variety that is showy inside and out. This fruit is large, ranging in size from 0.75 – 1.5 lbs (0.34-0.68 kg). No cross or hand pollination is needed. This variety is undoubtedly worthy of attention by commercial growers and homeowners alike.
10. **Purple Haze Dragon Fruit (*Hylocereus undatus*)** – Purple haze is a large sweet fruit with relatively few seeds. It weighs up to two pounds, and they have a pleasant grape, kiwi-alike flavor. This variety is self-pollinating and, like all other magenta-fleshed fruit, does not need other varieties (pollinators) to produce fruits. This variety has commercial value and can be eaten fresh, in juices, or used as a garnish.
11. **Red Jaina Dragon Fruit (*Hylocereus polyrhizus*)** – Of the varieties, we have at P.I.N. (Pine Island Nursery), this one is the heaviest producer thus far. The fruit is typically 0.5-0.75 lbs (0.23-0.34 kg) with especially dark red flesh. It is semi-sweet and good to eat, but its greatest potential is as a juice variety. The crimson red lycopene-enriched juice could compliment a multitude of tropical drinks, smoothies, or culinary creations. This variety is not self-pollinating and needs to be crossed with another to set fruit.
12. **Seoul Kitchen Dragon Fruit (*Hylocereus undatus*)** – Seoul Kitchen is a medium to large fruit typically weighing 0.75-1.0 lbs (0.34-0.45 kg). The pulp is smooth and sweet, and it is excellent when chilled and eaten out of hand. This variety is self-pollinating, and it will set fruit without hand pollination.
13. **Thompson Dragon Fruit (*Hylocereus undatus*)** – Thompson is another selection made by California rare fruit growers. It is a large, robust fruit weighing up to 1.5 pounds (0.68 kg). The pulp is sweet and flavorful, and it is self-pollinating.
14. **Vietnamese Jaira Dragon Fruit** – Vietnam exports more fruits of this variety. The fruit is large, typically weighing 0.5-1.5 lbs (0.23-0.68 kg). It has a sweet snowy flesh, and its impressive size makes this variety so popular among growers and consumers worldwide. This variety is self-pollinating and will set fruit without being hand pollinated.
15. **Voodoo Child Dragon Fruit (*Hylocereus polyrhizus*)** – A fruit of a small size, Voodoo Child Dragon is sangria-colored and exceptionally tasteful. The fruit, which is greater than egg, has a sweet, grape-like taste. It is advised that the fruit be enjoyed from the fridge fresh or used for juice production, typically mixed with other juices. This variety is self-pollinating.

- 16. Yellow Dragon Fruit (*Hylocereus megalanthus*)** – Yellow fruit and whitish flesh is also known as Pitaya amarilloar and is the only species of the genus Selenicerious. It is quite different than the Hylocereus in color and harvesting time, with thorns often present on the fruit. The fruit is small to medium at 0.5-0.75 lbs (0.23-0.34 kg) and ripens from late November through February. This fruit is certainly one of the sweetest and most tasteful Dragon Fruit . This variety is self-pollinating, and fruit will set on its own.
- 17. Zamorano Dragon Fruit (*Hylocereus polyrhizus*)** – This variety was collected from the Agriculture University in Honduras, from which it took its name. It is a medium to large-sized fruit, typically weighing 0.65-1.0 lbs (0.3-0.45). The flesh has a sweet, mild flavor and is incredibly dark red. It is best to hand pollinate for larger fruit, but this variety will set fruit on its own. This fruit is versaiter and can be eaten as fresh fruit, made into juice, or used as a garnish.
- 18. Pitaya rojaor red-fleshed pitahaya (*Hylocereus costaricensis*):** Red fruit with red-pink flesh

5. Morphological Characteristics:

Dragon Fruit is easily recognizable due to its unique appearance and juicy flesh, which display various morphological features that define its distinct identity. The plant is typically a climbing or sprawling epiphytic cactus with segmented stems that can be green or reddish-purple in color. These stems often have aerial roots for support and water absorption. They may have three- or four-winged ribs with sharp spines along the edges, serving as both protection and structural reinforcement. Spiral button-like structures at the stem margins eventually develop into flower buds. The flowers bloom at night, boasting large, colorful blossoms with creamy-white interiors and vibrant exteriors ranging from greenish-yellow to pink or red, often with purple pigmentation. The fruits of Dragon Fruit are renowned for their diverse shapes, sizes, and colors, ranging from pink to red or yellow, with corresponding hues in the pulp. The leathery outer skin of the fruit is often covered in scales or bracts, containing numerous small, black seeds within the sweet, juicy flesh. These edible seeds contribute to the overall texture of dragon fruit, making it a visually appealing and delicious fruit with a unique taste.

6. Soil Conditions:

Dragon Fruit cultivation is successful in various soil types, underscoring the significance of well-drained substrates for optimal growth. Although Dragon Fruit plants can withstand different soil compositions, such as clay loam, they demonstrate the highest growth and yield in sandy loam soils enriched with organic matter. Sandy loam soils with effective drainage systems create an ideal setting for Dragon Fruit cultivation, facilitating root development and nutrient absorption. Nevertheless, it is crucial to avoid waterlogged conditions, as Dragon Fruit, like all succulents, is vulnerable to excessive moisture, which can result in root rot and impede growth. In dense, waterlogged soils, farmers can enhance drainage by integrating sand or small stone gravel into the soil mixture to boost permeability and prevent waterlogging.

An ideal pH range for growing Dragon Fruit is 5.5 to 6.5, where the soil is slightly acidic in type. This pH nature plays a critical role in enhancing the availability and uptake of essential

nutrients for the crop. Such ideal qualities abound in nature-enriched, sandy loam soils that have high organic matter contents, thereby creating a good environment for the crop. However, while clay loams can support and sustain the growth of Dragon Fruit, these soils can in many cases be too tight and hold more water than what is required. This would thereby cause the rooting system to be suffocated and nutrients leached out.

However, farmers can correct this by bringing in amendments into the clay soil, such as sand and gravel, which must have good drainage. This is done to do away with waterlogging tendencies and to create suitable conditions for the growth of the Dragon Fruit. However, with highly sandy soils, watering and fertilization frequencies become more due to the leaching out of water and nutrients. This calls for the need for soil management practices that are site and soil-specific to result in best chances for growing Dragon Fruit plants. This way, farmers will be able to serve the purpose of growing Dragon Fruit to make use of the highest amount in production.

7. Climate Conditions:

Dragon Fruit is suited perfectly for regions like Karnataka, Kerala, Tamil Nadu, Maharashtra, Gujarat, Odisha, West Bengal, Andhra Pradesh, and the Andaman & Nicobar Islands: this is an excellent growth area for this cactus. The required temperature is 20 to 30 °C. Draconian Fruit grows well even when there's a range of 38-40 °C or down to 0° C for specific periods. Beyond 40° C, the stem will be yellow, or injuries will be done to the same. Dragon Fruit plants are very sensitive to temperature changes away from this range, adverse effects harm the plant. Despite being a crop that requires good watering, Dragon fruit will not thrive in areas with heavy rain as they undergo flower and fruit drop. The Optimum annual rainfall for the cultivation of the Dragon Fruit is 100 - 150 cm yearly. End. Healthy growth of plants is promoted by well-drained substrates and sandy loam soils high in organic material, underlining the significance of soil quality in successful plant growth.

In contrast to desert cacti, Dragon Fruit, despite being classified as a cactus, requires ample water because it comes from tropical rainforests. This trait makes it appropriate for growing in almost all regions of India, except for places with low precipitation. Dragon Fruit needs between 1145 and 2540 mm of rainfall annually, showing how crucial sufficient moisture is for its development and fruit production. Studies evaluating the crop's adaptability to dry tropical climates in Coorg, Karnataka, emphasize the importance of meeting temperature and water needs. On the other hand, regions with high levels of rainfall present difficulties because too much rain can cause flowers and fruits to fall off, which can reduce the overall yield. Hence, it is crucial to carefully assess climatic elements like temperature and rainfall when cultivating Dragon Fruit. The focus should be on choosing suitable locations and adopting proper management strategies to minimize the impact of unfavorable weather conditions.

8. Site selection:

The process of choosing a location for planting and getting the land ready is crucial for successful Dragon Fruit cultivation, providing the best conditions for plant growth. Pitaya plants need well-prepared land for proper growth, as their shallow root systems are focused in the top 15–30 cm of soil. This includes breaking up the soil until it is fine, getting rid of weeds, and

making sure there is proper drainage. Using organic compost improves soil fertility by providing important nutrients for plant development.

Prior to planting Dragon Fruit seedlings, it is important to evaluate the prevalent pests and diseases in the region to establish efficient pre-control methods. Taking care of pest and disease control beforehand can prevent future decreases in yield and promote healthy plant growth. It is recommended to plant seedlings in pits that have been properly prepared, with a depth and width of around 0.6 meters, at least one month before planting. The square plantation system is frequently employed on level terrain, maximizing space usage, and streamlining management procedures.

When choosing a spot to plant Dragon Fruit, multiple factors need to be considered. The incline of the land, its height, and the direction of rainwater are key factors in identifying appropriate areas for planting. Dragon Fruit grows best in well-draining soil, such as sandy soil blends or non-sandy soil blends that have been enhanced with organic material. The planting holes need to be a minimum of 1-2 inches deep to fit the seedlings and encourage root growth. Through a thorough assessment of these factors and proper land preparation, farmers can establish optimal conditions for the successful cultivation of Dragon Fruit, increasing yield potential and promoting strong plant growth.

9. Propagation Techniques

9.1 Sexual Method of Propagation:

Dragon fruit seed propagation entails the collection, preparation, sowing, and germination of seeds, followed by transplantation. Let us go through all these phases in excellent detail:

9.1.1 Seed Propagation: Dragon fruit can be grown through seeding, but vegetative propagation is more common. Dragon fruit seed propagation involves harvesting seeds from ripe fruits and processing them for germination.

9.1.2 Seed Collection and Preparation: Collect seeds from ripe dragon fruits and separate them from the flesh. To clean the seeds thoroughly, wash them multiple times to remove all traces of pulp and any soil adhering to them. Cleaned seeds are then dried properly in the air.

9.1.3 Sowing and Germination: Once dry, the seeds should be sown in a well-draining yet moist potting mix. Plant each seed in its own container or cell in a seeding tray, about 1/4 inch deep. Ensure the soil remains consistently damp without becoming overly saturated. Place the containers in a sunny spot for optimal germination conditions. Germination usually takes place in 2-4 weeks, with timing varying based on the environment and seed quality. During this period, ensure the soil stays damp and provides proper airflow to prevent fungal infections.

9.1.4 Transplanting Seedlings: Once the seedlings have developed multiple true leaves and are large enough, transfer them to larger pots or plant them directly outside in the soil. They thrive in locations with good drainage and plenty of sunlight and should be planted with enough space between each one. When transferring seedlings, handle them gently, carefully removing them from the pot without disturbing the roots. Place the seedlings at the desired distance apart,

making sure they remain upright and their roots are properly covered by soil. Water the seedlings adequately after transplanting to help them adjust to their new environment.

9.1.5 Care and Maintenance: Ensure the young plants receive enough water, especially during dry periods. Protect them from extreme temperatures and strong winds. Use a trellis or sticks to support their growth in the desired direction. By providing attention and care, a thriving member of your garden will grow and eventually bear fruits. However, seed propagation can result in variations among plants, so commercial farming often utilizes vegetative propagation techniques like stem cuttings to maintain specific characteristics.

9.2 Asexual/Vegetative Method of Propagation:

Dragon fruit is mainly propagated through cuttings. It could also develop from seeds. This method is not applicable in commercial farming because the seeds last longer and do not acquire the same attributes as the mother plant. Cuttings for planting must be procured from healthy plants. Cuttings suitable for planting in a field should be approximately 20 cm long. Pile up the cuttings two days before planting. Then, into these pots, the said cuttings should be put with the planting mixture of dry cow dung: Top dirt as 1:1:2. A week before planting, pots should be held in a shade. Maintain a 2 x 2 m space between individual plants. Holes should be 60 x 60 x 60 cm. Fill these holes with top soil and compost mixed with 100 grams of superphosphate.

Generally, the process involves the following steps:

9.2.1 Site Selection: Dragon fruit needs full sun and adequate drainage, so choose a sunny spot in your garden or a sunny windowsill that receives at least six hours of sun a day.

9.2.2 Cutting selection: Remove a 12-inch branch from a 3-year-old, fully developed plant. Prune an old pitaya plant with garden shears. Do not prune the plant too much; this will slow its growth.

9.2.3 Preparation for Cutting: Cut the pitahaya into three to five pieces. Each of these pieces can be used to grow a new dragon fruit plant. Make sure you remember which way is "up" for each cutting. When you plant them, they need to be planted upright for them to grow correctly. Using fungicide on each cutting can help prevent infection, but it's not necessary.

9.2.4 Healing: Put the cuts in a warm, dry place for a while so the edges will callus. The cuttings are ready when the tips turn white, which takes several days to a week.

9.2.5 Planting: To plant each cutting, set the base an inch or two below the ground and push the soil around to keep it from falling over. Make sure that the cutting is planted in the same direction that the original branch was growing. So the end that is closest to the base of the original dragon fruit plant goes down into the soil, and the end near the tip of the original branch points out of the soil.

9.2.6 Watering: Always water or spray the bed of soil so that the soil is slightly dampened. In three or four weeks, you should begin to see the shoots and roots start to enlarge.

9.2.7 Transplanting: Grow cuttings inside and transfer them, as they outgrow the pots, to a larger pot or a garden bed with the right climate.

9.2.8 Staking: Once your pitahaya has reached 12 inches, it will have to be supported to acquire height for further growth. After all, dragon fruit is like a climbing cactus. Place a trellis or a wooden stick on which your plant can grow.

9.3) Grafting Techniques

A horticultural process by which tissues of two unlike plants are joined in order to produce a hybrid doing well in the both good qualities. An example includes a cutting from a preferred type of dragon fruit that gets grafted onto the rootstock of another dragon fruit. All these induce better characteristics of resistance coming from the rootstock or robust growth on a grafted plant.

For the plant of dragon fruit, grafting is an important process since the bisexual flowers produce kinds of fruits that are excellent and which much could not be got either through the use of seeds or vegetative parts. Besides, it could also increase effectiveness, which means that this will lead to plant growth, which could be promoted. Also, grafting can increase vitality, and more so, it benefits the farmer further in investment to enlist a higher amount of yield and quality.

9.3.1 Benefits of Grafting Dragon Fruits

The grafted dragon fruit has many benefits, the major part of which lies in developing superior varieties of the plant, which would seldom be available in the market. Grafted dragon fruits are also tolerant to forbidding growth conditions due to better resistance to diseases and pests. Subsequently, better survival prospects translate into a greater final yield from the plantlets compared with that from seeding and cutting methods.

Grafted dragon fruits are vigorous, develop early into fruit, and exhibit rapid growth, thus quick returns on investment due to fast maturity of the succulent produce. In addition, grafted plants enter the fruiting stage early, show increased crop yields, and improve produce quality as a result of positive traits endowed by the scion and rootstock. In this regard, fruit growers are expected to see the benefits of grafting and prefer their plant material grafted.

9.3.2 Types of Grafting:

There are various grafting techniques conducted on dragon fruit, based on a different type of scenarios and considerations. In dragon fruit there are three most popular grafting techniques and they are explained below:

9.3.2.1 Cleft grafting: In this grafting techniques, vertical cut has created on rootstock and healthy selected scion is inserted in the cleft. It is a very simple and successful method. This is a very common method for larger rootstocks and is most suitable for beginners because it can give very high percentage success.

9.3.2.2 Whip-and-Tongue Grafting: This grafting procedure is pretty sophisticated and involves nice execution upon the scion and rootstock in a way that the two interlock in such a fashion that a tight union is formed. Because small rootstocks are used, this makes it a much more successful approach than cleft grafting.

9.3.2.3 Side-Veneer Grafting (or Inlay Grafting): A slant cut is made in both the rootstock and scion; thereafter, the scion is attached to the side of the rootstock to make a slanting union. Useful for large-sized rootstocks; the number of grafts possible per plant is several.

9.3.3 Procedure of Grafting:

Even at the mention of grafting dragon fruit, it does seem overwhelming, yet it is only very rewarding with proper preparation and actual technique. Below are steps on how to go about grafting of a dragon fruit:

1. **Selecting the Scion:** One should select a healthy shoot free from diseases of the desired variety of dragon fruit. The length of the scion has to be 6-8 inches with 3 nodes.
2. **Rootstock preparation:** Pick up a healthy, vigorous dragon fruit plant by the rootstock, taking off the side branches or else leaves, to get a clean straight section on the stem.
3. **Making the Cuts:** Take a sharp, sterilized grafting knife. Make diagonal cuts on the rootstock. Make a corresponding diagonal cut on the scion, ensuring that the notches fit perfectly together.
4. **Insert the Scion into the Cut over the Rootstock:** Insert the scion properly into the cut made over the rootstock. The union should be tight and proper, and cambium from either side should touch each other.
5. **Tying the Graft Union:** To secure the graft from heavy wind and storm conditions, the graft union should be tied by grafting or a rubber band. The band should not be tied too tightly, as that may create a blockage for the flow of sap.
6. **Aftercare:** Place the grafted plant in a warm, humid area; ensure that the soil around the plant is moist at all times; and watch for infection and graft union failure.

With these simple steps, dragon fruit grafting could be made a very successful process in unlocking the potential of these remarkable plants.

9.3.4 Major issues of dragon fruit grafting and the corresponding solutions

Grafting dragon fruit can be a very tricky job, particularly for beginners. Proper knowledge and techniques can tackle these challenges most efficiently. Some of the problems that one may face during grafting are mentioned here, along with their solutions:

- **Poor Union Forming:** There may be a case where no union is formed since the graft does not take hold together or is just a weak one or a total failure. Again in this situation, correct union can be made by cutting both the scion and rootstock with very clean, extremely sharp tools whereby the scion is cut of the same diameter as the rootstock for better fitting.
- **Infections and Diseases:** Grafted dragon fruit plants will also begin to be infected and affected by diseases, primarily during the initial stages of grafting inauguration. During grafting, it would be great to observe a clean environment, use sterile equipment, and avoid having bare hands touching cut surfaces to prevent this.
- **Graft Rejection:** Sometimes, the scion fails to pick up from the rootstock due to various reasons. One of these is the poor compatibility of varieties, while another is simple poor compatibility between a scion and rootstock. Graft rejection can be avoided by the use of

compatible varieties and ensuring the cambium layers of the scion and inceptor stock come into contact.

It can, however, be achieved by being patient and persevering, the end point being successful dragon fruit planting materials.

9.3.5 Caring for Grafted Dragon Fruit Plants:

When all the successful grafting is done, the provision for the plants is practiced, hence their good health and production. Important care tips for the plants are as follows:

- 1. Watering:** These plants require water consistently daily and, most of all, during the growing season. Let the soil be kept consistently moist but not waterlogged. Do not overwater because it results in rotting roots and other fungal diseases.
- 2. Fertilizing:** Use a balanced fertilizer purposely meant for dragon fruit plants. Apply as per the manufacturer's instructions on dosage and frequency. Caution, over-fertilization may result in high vegetative growth at the expense of fruit.
- 3. Amount of Sunlight:** Dragon fruit plants need full sunlight to thrive. Ensure that your grafted plants obtain at least six to eight hours of direct sunlight daily. Use shadings during extremely hot days of summer to protect from sunburn.
- 4. Pruning:** Regular pruning should be done with your grafted dragon fruits. Trim off dead or crooked branches with diseases and excess growth. Pruning encourages a better aeration system and lessens the possibility of fungal diseases.

With these care tips, you can be sure of having your grafted dragon fruits grow into and be very productive.

10 Planting and Spacing

In order for dragon fruit to maximize growth and high production, site preparation, planting style or method, trellising, and judicious planting spacing are very important.

10.1 Site Preparation: We will dig the land well by using a tractor before planting. Ploughing will be done until the soil attains fine tilth and is free from weeds. The land should be allowed proper drainage, and the land area should be fed with organic dungs and din to improve the fertility of the land and support better growth of the plant. Dragon fruit plants grow and produce good quality fruits under good sunlight conditions, i.e. a minimum of 6-8 hours of sunlight every day. The vegetative material should not be planted in shadowed areas, and shadowing of one plant by the other should be avoided. Maximize light penetration into the crop by selecting the optimal planting distances.

10.2 Planting Technologies

Transplanting is best done a month before planting into well-prepared pits whose sizes are 0.6m in both width and depth. In flat lands, the square system of plantation is used. Planting is done close to support poles. Initially after establishment, immature stems need to be tied to the support structures until the aerial roots develop and naturally tie onto them.

10.3 Support Structures

Support structures are crucial for the upright growth and productivity of dragon fruit plants. These structures help the plants to mature and produce lush fruits efficiently. Here are the key aspects of support structures for dragon fruit cultivation:

- **Types of Support Structures:** Dragon fruit plants require robust support due to their rapid and heavy growth. Commonly used support structures include stone or concrete poles, which are initially used to tie immature stems until they develop aerial roots and bind naturally. Trellis systems, such as single poles or wires stretched across rows, are also effective. Single-pole trellises are particularly beneficial for commercial production, supporting the plant's weight and facilitating easy access to flowers and fruit.
- **Material and Construction:** Support columns should be made of durable materials like concrete or wood, as fully grown plants with aerial roots can be very heavy. A round metal or concrete frame around the plant helps maintain balance. The recommended dimensions for support poles or trellises are 4.7 inches (12 cm) in diameter and 6.6 feet (2 meters) long, with at least 1.4 to 1.5 meters (4.6-5 ft) above ground level after installation.
- **Spacing and Arrangement:** Provide a circular metal frame for better support and allocate 8×8 feet (2.4×2.4 m) of space around each plant to allow uniform growth. The supporting poles or trellises should be arranged to maximize light penetration and minimize shading between plants.
- **Effective Trellis Systems:** Single pole trellises, continuous pyramid stands, and 'T' stands with two different cultivars are some of the trellis systems used. Among these, single pole trellises are proven to be the most effective for commercial production, supporting plant weight and allowing easy access to flowers and fruit.

By utilizing these support structures, you can ensure the robust growth and high yield of your dragon fruit plants.

10.4 Optimal Plant Spacing: Good spacing will prevent overcrowding, allow for good air circulation, and thereby allow maximum light penetration for the dragon fruit plants. Consider planting dragon fruit plants at least 15-25 feet from a tree, structure, or electrical line to give room for their spread. Recommended spacing patterns range from 10-16 feet between plants and rows. Closer spacing accelerates production, in contrast to wider spacing, which makes the plants more susceptible to diseases and allows air circulation. This becomes greatly important for small-scale farmers. The type of support post used influences the spacing. For single posts, 3 m x 3 m, plants are usually spaced at intervals. In general, each pole supports 2-4 plants, hence the length of the pole is 1.5 m - 2 m; it usually depends on the micro-climatic conditions. To get a well-balanced shrub, with increased fruit-bearing, limit the number of the lateral shoots up to 2-3 main stems. Adequate spacing will prevent the plant from falling over and ensure robust growth. Dragon fruit plants grow vigorously in full sunlight, so the best place to plant them is in open areas, and not in the shade. Not at all, in shady spots, so that these are not to be let away from growth or yield maximization. In so doing, with the consideration of all the factors, there will be

a good planting environment and planting of dragon fruits, which will bring the best growth conditions and yield.

11. Plant Density: Appropriate spacing should be provided for around each dragon fruit plant so that it leaves enough space for the plant to spread out after growth, without the interference of trees, structures, or electrical lines within 15-25 feet (4.6-7.6 m) distance. Adequate spacing to the plant helps in preventing shadowing and obstructing resources for the plant. Plant stands can range from 10 to 16 feet apart within rows, with the growth aspects and production optimized.

Spacing (m or feet)		Number of plants per ha or acre
Between rows	Within rows	
3 or 10	3 or 10	1100 or 446
4 or 13	3 or 10	833 or 337
4.9 or 16	3 or 10	680 or 275
3.7 or 12	3.7 or 12	730 or 296

12. Dragon Fruit Pollination:

Pollination of the dragon fruit is very important for fruit production, especially the self-incompatible varieties that need cross-pollination among different varieties to ensure better fruit set and size. To maximize potential for fruit production, farmers are advised to plant 2 to 3 different varieties (not the same clone). Examples of dragon fruit flowers are such that they are nicknamed the "lady of the night," which usually open during dark hours for pollination, such as with moths, hawks, and bats. Some varieties have flowers that last until early to mid-morning, which allows cross-pollination by bees. However, most of the countries where the dragon fruit is a new crop, having good natural pollinators, lack and therefore give poor fruit set to dragon fruit. Hand pollination also offers an effective substitute to natural methods, which often serves to improve fruit set. This process involves the collection of pollen from a flower and its application on to another flower's stigma; ideally this is done at night or early in the morning. Dragon fruit varieties such as American Beauty, Dark Star, Delight, Florida Red, Giant Vietnamese, Hailey's Conet, Makisupa, Rixford, Seoul Kitchen, Surinam Red, Thai Dragon, Thomson, Vietnamese White, Voodoo Child, Wiangel, Condor, Natural Mystic, and Purple Haze are known for their self-pollinating capabilities. These varieties ensure reliable fruit set without requiring cross-pollination from different plants. Each variety offers unique characteristics in terms of fruit size, flavor, and appearance, catering to diverse preferences among growers and consumers alike.

13. Training for Dragon Fruit:

Training Systems: Since the Dragon fruit belongs to the group of vigorously growing vines, they first form dense thick branches. Hence, they need to be trained properly, leading them towards proper growth.

13.1 Structural Pruning for Trellis Formation : In early growth stages, it is recommended to remove lateral buds and branches to promote vertical growth towards the trellis. After having reached the top of the trellis, the branches should be allowed to grow freely. On tip-pruned main stem, lateral shoot growth promotes a plant's umbrella structure on the trellis. This umbrella

structure helps in the support of flower development and fruit production by causing lateral branching. Dragonfruit vines can bear 30 to 50 branches in a year, and within four years of full-grown vines, they can overtake 100 branches.

13.2 Evaluation of Several designs of trellis in India: Several designs of trellis have been standardized in India like single poles with cement and iron ring, continuous pyramid stands, 'T' stands with different cultivars. A study conducted by IIHR Bengaluru stated that the single pole system with ring type trellising showed significantly better growth and yield performance compared to other systems.



(Source: telanganatoday.com)

13.3 Commercial Viability: The single pole with ring type trellis is proved to be more apt for commercial mode of cultivation. It held plant loads effectively and allowed flowers and fruit to be easily accessed, therefore speeding up the harvesting process. Wooden poles are rough to use, and are less lasting than cement poles. It is also impractical to change them once the vines have twined around them. So, despite the greater initial cost, one recommends the use of concrete poles because they will be more durable and permanent when used in dragon fruit.

14 Dragon Fruit Pruning

It can be very destructive in the growth and productivity of the dragon fruit when done through selective removal of parts such as branches, buds, and spent flowers. Without this measure, dragon fruit is a vigorous grower and will become rampant without much effort. It is therefore an overall necessity to carry out the procedure of cutting to maintain shape and size for the best growth and yields. One good vine of dragon fruit can have up to 30 branches in a year. Pruning also provides convenience for shaping the plant and easy access for harvest. Proper disposal of the cut parts, then, is critical for suppressing their growth into weeds and hampering the crop's growth.

14.1 Pruning Steps of Dragon Fruit Plant

- 1) Timing:** Pruning can be carried out after the last harvest – in May, or June
- 2) Cleaning of Main Stem:** Remove branches growing in main stem

- 3) **Removal of Old Growth:** Eliminate some of the old branches selectively, which grow from under the canopy
- 4) **Cutting Precisely:** Backward cutting of branches to the original stem can be done properly
- 5) **Canopy Size Establishment:** One to three sub-branches for every main branch should be maintained and remaining cut away.
- 6) **Future Maintenance:** This will require regular pruning to encourage fruiting to an optimum for October to flower in September to October.

Here are the branches one dragon fruit plant has on average over the year:

(Age) in Years	No of Stems per Plant
1	30
2	70
3	100
4	130
5-6	150-170

14.2 Do's and Don'ts in Pruning:

- 1) **Select Wisely:** When one is pruning, it's best to choose the lighter green branches and leave the darker green ones intact. These are usually older ones and can bear fruit. When it is a fruiting year, trimming the tip of the branch will make the plant produce more branches from that single stem.
- 2) **Regular Pruning Schedule:** Generally, young dragon fruit plants are pruned once in a growing season, and mature plants 2-3 times a year. Heavy or main pruning is done right after the harvest of fruits wherein dead, tangled, or non-productive stems are removed.
- 3) **Plant Canopy Structure:** Overcrowding of plants may lead to reduced photosynthesis and plant growth due to the shading effect. Tangled stems have minimal aeration that results in more disease infestations. Good pruning refreshes the plant as it induces flowering and fruiting.
- 4) **Not Pruning:** Avoid letting them grow at their will. This is because the excessive growth brings about too much shade within the canopy, thus, it leads to poor growth and fruiting.

14.3 Pruning of Dragon Fruits

1. **Training Pruning:** This is commenced twenty-three weeks into plantation where all shoots but one or two are removed. These are allowed to go up to the end of the support structure. Lateral shoots are continuously removed to the top to cause formation of lateral shoots from the terminal end.
2. **Sanitary Pruning:** It consists of eliminating those pods attacked by pests or diseased and shoots situated awkwardly. The cuts are made at the level of the internodes. Infected material is discarded outside the farm to prevent the spread of disease and other problems. Side shoots throughout their growth are removed, allowing only the main shoot until it reaches the desired length.

3. Production Pruning: These always strike a balance in vegetative growth whereby the harmed, dead, or very long stem is removed. Also, it makes the crown lighter unlike overgrown which makes harvesting of fruits tough.

14.4 Advantages of Pruning

- 1) Regulate the plant's vigor to enhance the number and quality of the fruits.
- 2) Simplify manipulation since it enables maintenance of a suitable size and form.
- 3) Sick, damaged, or dry branches are cut for the general benefit or health of the plant.
- 4) The branches that are pruned can be used for propagation so that there will be no break in planting.

15. Water Requirement and Irrigation Systems

Dragon fruit is succulent plant hence more resilient to drought and can flourish in low water. Adequate soil moisture, however, is must to achieving the greatest growth and performance of a plant. By and large, the water requirements of a pitahaya are near that of a papaya. One popular practice employed by farmers involves depriving plants of water immediately prior to flowering to encourage flowering. Good irrigation is essential during fertilizing and for flower and fruit development. The soil should be allowed to dry before irrigation again to avoid rotting after fertilization has been added. After potting, it is recommended to water gradually and make sure the water drains out at the bottom before throwing out extra water to avoid accumulation. If the soil is allowed to dry too much and the plant is not watered often, this can lead to increased fruit splitting.

It is highly advised to water potted dragon fruits at their roots to make sure that the roots consume only the necessary amount of moisture. The extra quantity will flow out to prevent excessive watering or root rot. Under normal circumstances, it is recommended to water every two weeks. Give the soil a chance to be: Dry or slightly moist, never completely hardened, and never too wet to allow for proper aeration. We can analyze the soil's moisture level by using a moisture detector meter or using finger-touch sensation. In India, we water the Dragon Fruit plant based on the specific local environmental conditions. On average, the dragon fruit plant needs between 800 mm and 2,500 mm of water per year from either irrigation or rainfall to thrive. During the rainy season, additional watering may not be necessary. During the summer season, there may be a large amount of weekly water consumption, requiring multiple waterings each week. Dragon fruit plants cannot withstand saturated roots; excessive water from overwatering or heavy rainfall can lead to root rot. In areas with extremely high precipitation, it is crucial to carefully think about drainage.

Typically, depending on the plantation's size and workforce, pitahaya plants can be watered manually (using buckets or setting up an irrigation system). Generally, farmers nowadays mostly prefer micro-irrigation systems that provide higher efficiency. Aside from being more effective, these systems are usually less expensive than manual irrigation due to the necessity of employing numerous workers and spending numerous hours. The use of drip irrigation system is implemented as a new method of watering dragon fruit plants in farms worldwide. Water flows through a small tube and trickles down the plant, regulated by a drip

irrigation system. This is frequently utilized in the arid region of the country. The plants' water use efficiency can see a significant increase (up to 90%) with the implementation of drip irrigation, as it allows water to be added in smaller amounts and with more accuracy near the plants' active root systems. Simultaneously, this system decreases the runoff and evaporation of the soil water that is either applied or stored. Upon installation on a farm, it allows the farmer to choose to utilize fertigation, which involves the simultaneous application of fertilizers and water through the irrigation system.

The dragon plant is very tough against low rainfall and prolonged drought, but to have good quality in the fruit production, steady watering has to be followed. Consistent irrigation also ensures quality fruit development. Here, in the Andaman Islands, eight months have adequate rainfall, and hence, even if about six hours of drip irrigation in alternate days can be given during the rainy season to ensure that there are some soil moisture-retaining organisms in the dry months, irrigation may not be necessary in the remaining four months. Effective control of weeds with the use of weed mats not only suppresses the weed growth but also helps in the conservation of soil moisture, which is very important as the root spread of dragon fruit is generally shallow, 15 cm to 30 cm.

It is imperative that there is proper irrigation maintained to deliver enough water in the event of a dry spell. Effective service drainage in the rainy season is equally important to prevent waterlogging. Long periods without irrigation could record serious reductions in both fruit yield and quality, thus placing great importance on dry periods before flowering to enhance the capacity of producing fruit. Where it relates to localized drips for irrigation systems, it has been proven successful in the optimization of yield and growth. Water is delivered directly to the point it is needed, without allowing it to go to waste; a unique characteristic not found in flood methods, which are discouraged due to their inefficiency and increased effort to be implemented against weeds. While most plants' watering average is 2-4 liters twice a week during summers or the dry season, the herbage and water requirements vary in regard to type of soil, climatic conditions, and general health status of the plant. Irrigation practice as per necessity is vital for the desired growth and productivity of crops.

16. Fertilizer Management for Dragon Fruit

Soil Preparation and Analysis: Before starting commercial dragon fruit farming, it's crucial to perform soil analysis to determine existing and lacking nutrients. This allows farmers to meet the specific nutrient requirements of the pitaya plant.

Soil Treatment: Dragon fruit thrives in slightly acidic soil. To maintain optimal soil conditions, treat acidic soil with ferrous sulfate and treat basic soil with chelated iron.

16.1 Nutrient Requirements:

Dragon fruit plants need proper nutrients for better yield. Their root system efficiently absorbs even small quantities of nutrients. The recommended doses are 450:350:300 grams (N-P₂O₅-K₂O) per plant. The application schedule is divided into four doses:

- 1. Before flowering:** 10%, 10%, and 30% of the total N-P₂O₅-K₂O.
- 2. At fruit set:** 20%, 40%, and 25%.

3. At harvest: 30%, 20%, and 30%.

4. Two months after harvest: 40%, 30%, and 15%.

Meeting these nutrient requirements ensures optimal fruit weight, shape, and quality. Calcium and micronutrients can be applied to boost fruit growth and firmness. Low nitrogen fertilizers, such as Super Bloom (0-10-10 or 2-10-10), are also recommended.

16.2 Fertilizer Practices in Different Countries

1) Taiwan:

1. 4 kg of organic manure per plant every 4 months.
2. 100 g of commercial 13-13-13 NPK mixture per plant.

2) Hawaii

1. 180-230 g per plant of 16-16-16 NPK mixture every 4-6 months.
2. Calcium from crushed eggshells (90% Calcium, 6% Magnesium, and 1% Phosphorus).
3. Super Bloom (0-10-10 or 2-10-10).

3) Vietnam

1. Young plants (less than 3 years old): 10-15 kg of farm manure per plant.
2. At planting: 100 g of super phosphate per plant.
3. First two years: 300 g of urea and 200 g of 16-16-8 NPK per plant annually.
4. Mature plants (over 3 years old): 540 g N, 720 g P₂O₅, 300 g K₂O, and 20 kg of farmyard manure per plant annually.

4) Sri Lanka

1. NPK 1:1:2 at 30-40 g per vine, three times a year.
2. Alternatively, 100 g per plant annually (15 N – 5 P – 15 K – 8 S – 1.6 mg per gram).

5) Philippines

1. Apply combined complete fertilizer (14-14-14) and organic fertilizer every 3-6 months.
2. Recommended dose: 2 kg of organic fertilizer plus 25 g of urea and 75 g of complete fertilizer per plant.

6) India

1. Best results for yield and quality with N 450: P₂O₅ 350: K₂O 300.
2. Apply in four split doses per pillar with four plants
3. Before flowering: 10%, 10%, and 30%.
4. At fruit set: 20%, 40%, and 25%.
5. At harvest: 30%, 20%, and 30%.
6. After two months of harvest: 40%, 30%, and 15%.
7. Combine organic manure with neem cake and 100 g of 19-19-19 complete fertilizer every 3-4 months. Regularly prune plants to maintain an open and manageable canopy, promoting new shoots for the next cropping season.
8. Dragon fruit can be grown organically without inorganic fertilizers or pesticides, complying with the high international demand for organic-label fruits. Use organic manures such as cattle or poultry manure or well-decomposed compost to meet nutrient requirements and enhance market competitiveness.

17. Weed management: While there are no significant reports of weed problems in dragon fruit production, effective weed control is essential for healthy crop growth. Weeds can harbor diseases and pests that threaten fruit crops. Therefore, proper and consistent weed management is crucial to maintaining a healthy dragon fruit plantation.

18. Plant Protection of Dragon Fruits

With very little information available, the production of dragon fruit is gaining wide momentum around the world, and additional hectares get under its cultivation each year. The only limitation it establishes is through the pests and diseases that lower the plant's health and reduce the final output. This chapter discusses the main insect-pests and pathogens that lower the production of dragon fruits.

18.1 Major Pests of Dragon Fruits and Its Control

18.1.1 Oriental Fruit Fly (*Bactrocera dorsalis*): This pest originates from most tropical parts of Asia but has now been introduced in parts of sub-Saharan Africa and the USA, where it is found in Hawaii. It is the significant and hazardous pest that might destroy the crop terribly in case of not being controlled by the farmers. Also, its quarantine status implies, and post-harvest sanitization of the fruits that are to be exported is recommended. In most scenarios, the insect comes in immense numbers after a heavy downpour of rain. It is the activity of the larvae on the fruiting stage that causes the damage mainly. More accurately put, the female insect lays eggs in the skin of the fruit where they hatch and the larvae burrow into the fruit, causing either decaying or premature dropping.

Control Measures:

1. Bagging of fruits to prevent fly damage
2. Hanging methyl eugenol traps for long-term control
3. Spot spraying of pesticide and hydrolyzed protein food bait
4. Maintain field sanitation management of insect populations

18.1.2 Mealy Bugs (*Ferrisia virgata*): This pest is small with soft bodied structure covered with white powdery wax. It is cell sap sucking pest and excreta honeydew like sticky substances which attract ants.

Control measures:

1. Proper applications of pesticides
2. Liquid soap and soybean oil emulsions can be low-density infestation control

18.1.3 Ants: Ants can attack dragon fruit at any growth stage. They damage growth, shoots, buds, or fruits, which in turn will delay plant growth or even cause injury to plants.

Control measures:

1. Use of bait to regulate the population of ants
2. Spraying of soap or chlorpyrifos-based insecticide on affected parts of plants

18.1.4 Scale Insects: Scale insects attacked on surface of stems or branches.

Control measures:

1. Spraying of liquid soap mixed with water on the attacked parts

18.1.5 Flower Beetles: These beetles attack and spoil the reproductive parts of flowers, thus no fruits will be produced. The control on flower beetles includes;

Control measures:

1. Chemical control
2. Control of weeds that will regulate and restrict the populations of beetles

18.2 Significant Diseases of Dragon Fruit

There are several serious diseases that affect dragon fruit crops like anthracnose, brown spots, and stem rots caused by fungal and bacterial pathogens. Heavy rainfall, overwatering, or waterlogged conditions usually intensify these.

18.2.1 Fungus and Bacteria-induced Stem Rot: Stem rot is caused by either a bacterial infection *Xanthomonas campestris* and *Erwinia carotovora*. This is a disease that attacks the stem rotting and yellowing it; then, red-brown lesions appear and later turn whitish in the center.

Control measures:

1. Weekly spraying of Copper fungicide (copper oxychloride 0.2%), mancozeb, and/or metalaxyl products
2. Pruning infected plant parts and burning

18.2.2 Anthracnose (*Colletotrichum siamense*): Anthracnose due to the fungus, *Colletotrichum siamense*, has been reported to occur in India from the Andaman Islands. The causal organism induces the formation of reddish or orangish brown, concentric lesions bearing acervuli or black pinheads starting near the ribs of the vine, more frequently at the points of spine emergence. It also attacks the fruits.

Control measures:

1. Preventive spray: Chlorothalonil or mancozeb 2g/L.
2. Curative spray: Carbendazim 1g/L.

18.2.3 Stem and Fruit Canker (*Neoscytalidium dimidiatum*): This fungi disease is found widespread in South Florida and has been severely problematic in Southeast Asia, the Middle East, and North America. Sunken chlorotic spots on the stem enlarge to big convex orange to reddish-brown lesions. Control suggestions are as follows:

Control measures:

1. Maintain cleanliness to reduce pathogen populations
2. Remove and destroy diseased stems
3. Improve airflow and fungicide penetration into the canopy

18.2.4 Wilt Disease: Wilt disease caused by *Fusarium* species is drying or loss of turgidity of the plant. Unpublished data.

18.2.5 Rotting Diseases: Dragon fruit rotting diseases result from various fungal species such as *Alternaria*, *Bipolaris*, *Rhizopus*, and *Dothiorella*. Although these are not reported to be diseases suffered in India, it constitutes a major risk in other countries towards which the fruits are being transported.

18.2.6 Bacterial Rot: This is caused by *Xanthomonas campestris* and *Erwinia carotovora*. Heavy sunshine and calcium deficiency increase the susceptibility of plants to this disease. Sunburn enhances the symptoms. Management

18.3 Postharvest Disorders of Dragon Fruits

There are various postharvest disorders in dragon fruits that affect their quality as well as marketability.

18.3.1 Chilling injury: Chilling injury is also common when fruits are harvested early and this happens at 5-6°C. In this disorder, flesh translucency and softening will occur with wilting and darkening of scales. There is browning of the outer flesh with poor flavor.

18.3.2 Mechanical injury: Mechanical injury due to skin abrasion results in unattractive fruits with enhanced loss of water that leads to shriveling of fruits.

18.3.3 Moisture loss: Another related concern is moisture loss, as the fruit is prone to physiological weight loss ranging from around 0.1% at 5°C to 2.6% at 20°C. The problem can be alleviated by packaging appropriately in perforated plastic bags and ensuring the required relative humidity, as water and moisture loss result in unsightly shriveling and reduced fruit weight.

18.3.4 Decay: Decay is also one of the biggest threats to postharvest quality in dragon fruits.

18.3.5 Sunburn injuries: Also, sunburn injuries have been reported in most of the Indian plantations, especially during the months of March and April when the fluctuation in temperature is more and day temperatures are above 38°C. The symptoms of the same include discoloring and scorching of the fruit surface. In controlling sunburn injury, studies were done on the cultivation of dragon fruit under shade net houses, spraying antitranspirants and with filler crops to reduce physiological injury. It is for this reason that IIHR has taken steps to conduct experiments in off-season dragon fruit production.

19. Dragon Fruit Harvesting and Post Harvest Management:

19.1 Harvesting: The dragon fruit plants start producing after 12-15 months from planting. The fruits are mature when the epicarp color changes from green to red and are ready to be harvested seven days later. Fruits are produced from June until September, and harvests can be made three to four times a month. The weight of individual fruits varies from 300-800 g and the average yield of 30-35 kg per post for three year old plants. The present selling price at farm gate is INR 80.00 to 120.00 per kg.

19.2 Time and Method of Harvest: Dragon fruit is harvested 28-32 days after flowering. In the case of direct selling to consumers, harvesting is done 30 days after flowering, its scales still green. The dragon fruit plants produces fruits in late April or early May. Peak season is in June and July. Harvesting season ends in October or November and also in December in some areas, with 6-12 harvesting sessions or cycles in a year.

This is based on the quality characteristics as well as maturity indices such as skin color that turns to almost completely red. Harvesting at the wrong time can lead to poor quality, short storage life, and high susceptibility to chilling.

1.3 Steps to Harvest Dragon Fruit:

1. Choose a fruit which is not yet fully ripened or has turned pink as suggested by the variety.
2. Cut close to the top of the fruit, choosing the bigger fruits, in order to separate the fruit from the stem.
3. Place the harvested fruits in crates and proceed for storage or packaging.

19.4 Yield: The yield per post ranges from 20-35 kg depending on the variety, environmental conditions, cultivation practice, and age of the plant. The average yield per hectare ranges from 16,000 to 27,000 kg per hectare depending on planting distances, corresponding to 14,275-24,090 lb/ac.

19.5 Storage and Shelf Life:

The most optimal state for preserved dragon fruits is at 10°C with RH 93%, enabling the fruits to last for 15-17 days. If stored at too low a temperature, it turns soft, risking mechanical damage; if the temperature is high, the spine turns yellow, and the fruit no longer looks fresh. The appropriate cool technology should be maintained so that the fruit will remain fresh and high in quality during transportation. The dragon fruit is a non-climacteric perishable fruit that grows mold during storage. Published research showed that peppermint oil application diminished surface wounding, in addition to lengthening shelf life, as well as transmitting firmness, green bract color, titratable acid value, and total phenolic content up to 21 days compared to untreated fruit.

19.6 Packaging, marketing and exporting dragon fruits: The dragon fruit business seems to have a bright future both in local markets and export markets with peak global demand. Currently in 2022-2027 fruit market growth rate is 3.9% per annum. Dragon fruit is greatly gaining popularity in the U.S, although new to Europe, yet promising market and has attracted more consumers. In the Asia Pacific, it is the leader in production and consumption.

Packing Process

1. Receiving and Unloading at the Packing House: Fruits are unloaded from different farms, identified by area code, and recorded
2. Inspection, Selection, and Storage Product The fruits are sorted according to size, quality, variety, and classification. Size grades vary by country. For instance, Vietnam classifies dragon fruits as:
 - a. Extra-large: over 500 g (17 oz)
 - b. Large: 380-500 g (13.4-17 oz)
 - c. Regular: 300-380 g (10.6-13.4 oz)
 - d. Medium: 260-300 g (9.2-10.6 oz)
 - e. Small: less than 260 g (9.2 oz)

19.7 Fruit Processing After Harvest: Processing from the dragon fruit pulp and the juice processed with a solution containing 1.5% pectin, 55% sugar, and 0.9% citric acid improved significantly the colour and other organoleptic characters of jam and jelly from dragon fruit. In the dragon fruit RTS beverage, the most acceptable formulation was found to contain 14% pulp, 12% sugar, and 0.9% citric acid. These prepared products were organoleptically acceptable and could be stored for not less than three months at ambient conditions without microbial spoilage

or a considerable loss in quality. Dragon fruits are dipped in a sterilized water bath containing OZON detoxifying detergent for 5-8 minutes and then dried. The fruits are categorized by size and packed in 4 kg (8.8 lb) cardboard boxes or in 10 kg or 15 kg (22 or 33 lb) buckets, sealed with adhesive tape to prevent insect infiltration.

19.8 Unloading, Sealing, Shipping and Loading Containers: The dragon fruit boxes are loaded into reefer containers at the packing house. Shipping area is sealed securely to avoid return of products into the factory and also to avoid insect entry.

19.9 Factory Sanitation and Human Hygiene: There exists a disinfection and sanitation of all areas. The workers are also required in the measures to adhere to the hygiene. Records and logs of operations, customer feedback and periodic inspection are maintained for reference.

20. Import and Export:

Dragon fruit or Kamalam cultivation is rapidly gaining ground in India. Farmers of Karnataka, Kerala, Tamil Nadu, Maharashtra, Gujarat, Chhattisgarh, Odisha, West Bengal, Andhra Pradesh, Andaman & Nicobar Islands, Mizoram, and Nagaland take up its cultivation. The area under dragon fruit cultivation in the country is now more than 3,000 hectares. But the domestic production is not enough to meet the demand and hence there is heavy import of the fruit mainly from Thailand, Malaysia, Vietnam, and Sri Lanka.

Imports of Kamalam into India began in 2017 at 327 tons and then nearly shot up to 9,162 tons in 2019. The approximate imports for 2020 and 2021 were to the tune of 11,916 tons and 15,491 tons, respectively, while the value of import is put at approximately Rupees 100 crores for the year 2021. Dragon fruit is a short-gestation fruit because the commercial production begins in the first year, post culpa planting, and thereafter fully in 3-4 years. The life span of the crop is approximately 20 years. On an average, the plant will provide economic produce from the second year itself at 10 tonnes per acre. Considering the present selling rate of Rs 100 per kg, the annual return per acre would be to the tune of Rs 10,00,000, which works out to a Benefit-Cost Ratio of 2.58.

Recognizing it and in sync with the Atmanirbhar Bharat mission of reducing imports, enhancing domestic production capacity is imparted significance. Roadmap preparation about area expansion of dragon fruit has already been kick-started by Mission for Integrated Development of Horticulture for its cultivation in identified potential areas. It targets covering 50,000 hectares in five years. Plantations have recently been established at ICAR-Central Island Agricultural Research Institute, Port Blair, Andaman and Nicobar Islands, and IIHR, Bengaluru, Karnataka.

Under the MIDH Scheme, the Ministry of Agriculture & Farmers Welfare sanctioned one proposal for Centre of Excellence -Kamalam Fruit submitted by the Indian Institute of Horticultural Research, Hirehalli, Bengaluru, Karnataka on 09-03-2023. The area of focus for this centre will be Kamalam production, Postharvest and Value addition.

Centre of Excellence will develop latest production technology comparable to International level, help off-season production, and demonstrate technologies for high - yield production. It aims at self-sufficiency in Kamalam fruit production, value addition, and towards

the economic progress of the farming community. The Centre will undertake development of high performance varieties with enhanced yield, nutrient use efficiency, nutritional quality, and tolerance to biotic and abiotic stresses. Standardization of propagation techniques, supply of quality planting material through public participatory approach, and developing the protocols for post-harvest handling and storage for value addition, reduction of losses and promotion of exports. Further, the Centre shall focus on the development of value-added products and processes for diversified and value-added products and higher revenue accrual and also diffuse technologies to farmers and all other stakeholders through training and field visits.

Since more and more farmers are taking interest in it and with bumper returns from Kamalam cultivation in agricultural and marginal lands, new areas are likely to be brought under Kamalam cultivation and eventually substitute imports through domestic production.

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INFLUENCE OF ENVIRONMENTAL FACTORS (SOIL AND CLIMATIC) ON PERFORMANCE (UPTAKE, TRANSLOCATION, METABOLISM) OF SOIL APPLIED HERBICIDES

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Introduction to Soil Applied Herbicides:

Herbicide are applied directly to soil as pre-emergence spray or granules on the surface or incorporated into soil by shallow cultivation or injected below the soil surface. Herbicide once reach the soil, almost entire amount applied except that very small amount utilized for weed control, is subjected to various process of physical removal from soil and decomposition by several mean in soil and plants. A complex interaction operates between herbicide, plant, soil and climate.

Climatic and edaphic factors greatly affect the herbicide performance. All the process of physical removal neither alter the herbicides chemically nor to their deactivation completely. They just take herbicides away from the root zone of plants via leaching, run-off and vapourization almost permanently and through adsorption to soil colloids temporarily. The physical removal results in direct loss of herbicides in original form and with metabolites at various stages of decomposition based on availability of edaphic and climatic factors responsible for removal. Therefore, the process of physical removal may or may not operate instantaneously as soon as the herbicide is applied to soil since they are highly dependent on several soil and climate factors.

Example: if soil moisture or rainfall/water is not enough, physical removal of herbicide through adsorption, leaching or runoff may not be significant. The activity/effect of these process remains stationary/insignificant until there is enough rainfall or irrigation applied.

Photodecomposition when applicable to a herbicide, may operate instantaneously and does not depend on presence of soil moisture. Therefore, by the time when there was rain or field was irrigated, some amount of herbicide might have decomposed and formed metabolites and may be lost from soil along with parent materials.

Soil factors affecting herbicide performance (uptake, translocation and metabolism) in soil are: When herbicides applied to soil it is subjected to various processes of physical removal from soil and decomposition by several means, which ultimately determine their availability, phyto-toxicity, absorption and translocation. The physical removal involve Vapourization, run-off and wash off, leaching/deep percolation, adsorption etc which in turn affects herbicides availability in soil solution, by which they are absorbs by plant roots/seeds etc. Similarly soil

applied herbicide may undergo decomposition by microbial action or photo decomposition/chemical decomposition, which restrict their availability to target site. Following soil factors which affect herbicide action in soil are explained below:

Factors	Effect	Factors	Effect
Soil pH	Adsorption	Soil Texture	Adsorption
	Leaching		Leaching
	Microbial decomposition		Surface run-off
	Vapourization		Moisture holding
	chemical decomposition.		

Factor s	Effect	Factors	Effect
Soil moisture	• Absorption	Soil Organic matter	• Absorption
	• Activation of herbicide		• Phytotoxicity
	• Leaching		• Leaching
	• Adsorption		• Adsorption
	• Volatilization		• Volatilization
	• Microbial decomposition		• Microbial decomposition

Factors	Effect	Factors	Effect
Soil Temperature	Adsorption	Soil colloids	Adsorption
	Soil micro-organism		Vapourization
	Volatilization		Leaching
	Chemical decomposition.		

Sunlight : Photodecomposition

1. Effect of Soil pH on Herbicides Availability:

It plays major role in activity of soil applied herbicides. It affects several processes which in turn affect the herbicides availability in soil and thus its absorption.

A. Effect of Soil pH on Adsorption Herbicide: Adsorption is holding up of herbicides molecules, ions onto the surface of the soil colloids by physiochemical attraction. This leads to less availability of herbicides in the soil solution and in turn less uptake by plants. To be effective and safe pre-emergence herbicide must have properties that result in the majority of

herbicides being bound to the soil colloids with only a small amount remaining in the soil solution. If majority of applied herbicides remains in the soil solution the herbicide would rapidly leach through the soil profile or leave the field with runoff.

Soil pH can have a significant effect on adsorption of many herbicides. Some herbicides, principally triazines such as atrazine and simazine, are particularly affected by soil pH. Lesser amount of these herbicides are adsorbed or held to soil colloids at higher soil pH, so more amount is present in soil solution. Herbicides in the soil solution are available for plant uptake. As the pH decreases below 7 the concentration of hydrogen ion found in the soil solution increases. Many herbicides can incorporate hydrogen ion into their molecular structure, therefore tend to change the charge of herbicide molecule. e.g. at soil pH below 7, atrazine may pick up hydrogen ion from soil solution causing the atrazine to take a positive charge. The positive charge on atrazine molecule under acid conditions increases the attraction between herbicide molecule and negatively charged clay colloids. At soil pH above 7 most of atrazine maintains a neutral charge and thus the herbicide is less tightly adsorbed and more available to plants. Weak base herbicides, e.g. symmetrical triazines, triazoles, triazinines, thiazoles and pyridinones under acidic conditions accept a protonate each of their nitrogen atom to become a cationic species.

Weak acid herbicides (dinitrophenols, phenyl and phenoxy alkanic acid, sulfonyl urease etc) having carboxylic or phenolic-OH groups ionize in aqueous medium and yield their respective anions. The lower the pH, the lower the tendency for ionization and vice-versa. Adsorption of these herbicide through anion exchange is restricted mainly to non-silicate clay.

In case of some non-ionic herbicides, which do not ionize significantly in aqueous medium or soil analysis are dinitroanilines, anilides, phenyl carbamates, thiocarbamates and benzonitriles. Adsorption occur through hydrophobic attraction in case of non-polar non-ionic herbicides, whereas for polar non-ionic herbicides, adsorption occur in greater competition with water.

B. Effect of pH on Leaching of Herbicide: Leaching of herbicide through soil profiles depends on solubility of herbicide, amount of water, soil texture and adsorptive capacity of soil. It makes herbicide unavailable to target plants, while available to non-target plants or organism depending on how much leaching of herbicide has taken place.

Leaching is the vertical downward movement of herbicides with water through soil profiles to deeper layers and ultimately to ground water. The degree of leaching of herbicide is determined by adsorptive relationship between the herbicide and the soil, solubility of herbicide in water and amount of water passing downward through a soil.

Many solids exhibit a marked increase in solubility at both low and high pH values (amphoteric behaviour). Other solids may exhibit maximum solubility at neutral pH values. The pH of the leaching system before leaching occurs and described as the initial pH of the leachate. The leaching system pH at equilibrium is usually governed by the dissolution of the major soluble phases in the solid material. Frequently, the initial and equilibrium pH values differ

widely, particularly if the liquid to solid ratio (L/S) is low and solid phase dominates the system. When L/S ratio are high, the reverse can hold true; the pH of the initial solution is close to the equilibrium pH because the solution phase dominates. Except for the direct influence of pH on the dissolution, pH does also influence redox conditions, complexation and sorption.

C. Effect of pH on Microbial Decomposition: microbial decomposition is another major method of herbicide loss from soil, the rate of which depend on the environmental conditions and inherent nature of herbicide. soil pH also affects micro-organisms activity in soil as micro-organisms are highly influenced by soil pH. Bacteria and actinomycetes are active at moderate to higher pH and their activities decrease at acidic or lower pH. Soil pH lower than 4.5 drastically reduces their activities. Fungi, on contrary are less sensitive to soil pH and work well in almost all pH ranges of normal agricultural soils. They therefore, predominant in lower pH where bacteria and actinomycetes fall less active or inactive. When soil pH was raised from 5 to 7, soil microflora and degradation rate of EPTC increased and phytotoxicity was shortened 2 or 3 weeks.

D. Effect of pH on Vaporization: Soil pH affects vapourization of herbicides mainly by influencing their adsorption and favouring or disfavouring their dissociation. A low pH favour the undissociated form of anionic herbicides and increase their affinity towards vaporization. Soil colloid (clays and organic matter) content highly influences adsorption of herbicides and thereby their vaporization from soil.

E. Effect of pH on Chemical Decomposition of Herbicide: chemically breakdown of herbicides involves reaction such as hydrolysis, oxidation, reduction etc. e.g. in sulfonylurea (metsulfuron-methyl, sulfosulfuron and imazosulfuron) hydrolysis is major path way for its degradation in soil, which resulted into formation of sulphonamide and heterocyclic amine. Cleavage by hydrolysis is pH sensitive. As soil pH increases, the rate of chemical hydrolysis in soil decreases. Therefore, sulfonylurea herbicides degrade more rapidly at lower soil pH than at high soil pH.

2. Soil Moisture Content

Soil moisture is of prime importance to soil applied herbicides. It should be sufficient neither too much nor too low. Dry soil or excess moisture may cause failure of soil applied herbicides. Moisture help in incorporation of some soil applied herbicides with low solubility and increase availability for absorption by roots from soil solution. Soil moisture content play an important role in the herbicide absorption, transportation and metabolism as it affects the various physical and decomposition process of soil applied herbicides.

A. Effect of Soil Moisture Content on Absorption of Herbicide availability: If soil is wet and air is dry, plants transpire more. Roots absorb water from soil to replace transpired water and herbicides in soil move to roots by mass flow. More herbicide will be absorbed and phyto activity will increase. Some-times dry air and wind cause rapid foliar water loss and when not enough water is taken up by roots, plants wilt. Stomata then close, water movement in plants

decreases as a result of which herbicide uptake also decreases. Soil drying can increase soil adsorption and decrease root uptake.

The amount of herbicide in solution is directly related to the soil moisture content. In soils having already greater adsorption, irrigation may facilitate desorption of some reversibly adsorbed herbicide to soil solution even within available soil moisture range and may improve weed control, whereas in soils with lower adsorption, irrigation will dilute soil solution without inducing desorption of herbicide and may lead to poor weed control. The amount of space available for herbicides to go into solution decreases as soil dries out, thus less free herbicide is present in dry soils. Under dry conditions plants are exposed to less herbicide and therefore be less likely to absorb toxic herbicide concentration. The non-ionic herbicides adsorbed more on dry soil than on wet soil such as, EPTC. Moisture competes for sites on colloidal surface with the herbicides and induce desorption. The volatile compound thus desorbed are lost to atmosphere. When soil moisture replenished, herbicide will desorb from the colloids and re-enter the soil solution.

B. Effect of Soil Moisture Content on Activation of Herbicides: Rainfall or irrigation is essential to move herbicides into the top soil layers where most seed weed germinate. Some rain may be essential into activate herbicides such as the triazines, which is taken up by the roots. No moisture for 10 to 14 days after application can cause weed control failures.

Heavy rain, on other hand, may move herbicides below the zone of activity. Excess rainfall can leach herbicides through a zone of action unless they are adsorbed. The effect of soil moisture cannot be generalized for all herbicides, crops, or application times. Soil moisture play important role in several physical and decomposition process which affect the absorption, translocation and metabolism of applied herbicides.

C. Effect of Soil Moisture Content on Volatilization of Herbicide: The moisture content of soil has been determined to be one of the most significant environmental parameters which influence the rate of herbicide volatilization. Volatility generally increases with increasing temperature and soil moisture and with decreasing clay and organic matter content (Helling *et al.*, 1971). Volatility losses are greater when a herbicide is applied to a moist soil surface than a dry soil surface. Increased binding of a herbicide to a soil has been estimated to occur once the soil moisture content decrease to one to three molecular layers after the application on the surface of soil particle. However, the point at which increased herbicide binding occurs varies and is also depend upon soil composition which in turn, influences soil moisture capacity. The binding process is revertible and with increased soil moisture content such as after rainfall or dew fall, an increase in herbicide flux from the soil can be detected. Carbamates, thiocarbamates and dinitroanilines show direct relationship between moisture and volatilization. Herbicides bound tighter to dry soil, hence less volatilization. The volatility of non-ionic (EPTC) is higher in greater soil moisture as compared to lower soil moisture (Fang, 1969).

D. Effect of Soil Moisture Content on Microbial Decomposition of Herbicides: Soil moisture is very crucial for microbial activity. Excess moisture or water logging may lead to anaerobic

conditions and reduce the growth of aerobic micro-organisms. Similarly, in deficient moisture or dry soil, micro-organisms may die up or remain inactive/ dormant and as a result, the persistence of herbicides may be prolonged under moisture stress or dry soils. Microbial decomposition is optimum when soil is at or near field capacity. It however, takes place between 50-100% of field capacity.

E. Effect of Soil Moisture Content on Leaching of Herbicide: Soil water serve as a deriving agency in leaching of the herbicide. When there is high rainfall than the highly solubility herbicides along with water leached down to the deeper soil profile and may affect the non-target plants (crops). Leaching is primarily limited by rainfall and secondarily by soil compaction.

3. Soil Organic Matter (SOM)

Soil organic matter plays important role in different soil physical and decomposition process which in turn affect the absorption of herbicides by plants roots. Soil organic content is thought to be the best determinant of herbicide adsorption rates.

Effect of SOM on Adsorption of Herbicide: Adsorption of herbicides is correlated with organic matter and clay content and this effect is due to high adsorptive capacity of these soil constituents. The large surface area and chemical nature of humic substances can explain their high adsorptive properties. The organic matter content of soil therefore, influences initial phytotoxicity (higher adsorption, slow release in soil solution), volatilization (higher adsorption, less volatilization), leaching (higher adsorption, less leaching) and persistence (higher adsorption, less decomposition and thus more persistence) of herbicides.

Effect of SOM on Microbial Decomposition of Herbicide: Organic matter is source of energy for micro-organisms. If huge amount of organic matter is present in soil, it may result in increased herbicide adsorption. Once herbicide is absorbed then there is less chance for its decomposition by micro-organisms.

4. Soil Colloids (Inorganic and Organic):

Clay (inorganic aluminosilicate minerals) have lower adsorption than organic matter with similar surface area. Clay colloids play important role in adsorption when organic matter content is very low. Kaolinite (1:1) type clay minerals possess less adsorption than 2:1 types type clay. (Vermiculite>montmorillonite>illite>kaolinite).

Higher soil colloid content generally decreases the vapourization of herbicides, therefore soil colloidal content highly influences the adsorption and thereby vapourization.

5. Soil Texture:

Adsorption: Smaller the soil particles, greater is the surface area and therefore more adsorption. Clay will adsorb more herbicide than sand, therefore clayey soil have more adsorption than sandy soils.

Surface Run-off: Soil texture affects water infiltration rate, thereby affecting the amount of runoff. There tends to be greater runoff from finer-textured soils. Organic matter affects infiltration rate; there tends to be less water runoff as organic matter content increases. Organic matter also adsorbs herbicide, thus reducing amount of dissolved herbicide loss from the field.

Leaching: Coarse texture soil (sand) are more prone to leaching than fine soil (clay) because smaller the size, greater the surface area for adsorption and thus less amount of herbicide molecules are available for leaching.

6. Soil Temperature:

Effect of Soil Temperature on Adsorption of Herbicide: It hardly affects ionic adsorption, but physical attraction may fall weak to increasing temperatures. This is particularly pronounced for volatile herbicides and herbicides with negative heat of solution i.e. one which exhibit less solubility in soil solution at higher than at lower temperature e.g. thiocarbamate herbicides (Weber, 1972).

Effect of Soil Temperature on Volatilization: Volatilization losses increase as soil temperature increases. Atrazine and trifluralin, a herbicide having very low vapour pressure, may also volatilize if exposed to high temperature.

Effect of Soil Temperature on Soil micro-organisms: The optimum temperature for microbial activity is about 24°C- 32°C. Microbial activity is greatly diminished in cool temperatures.

Effect of Soil Temperature on Chemical decomposition: Chemical hydrolysis of sulfonylurea herbicides is fast during the summer when soil temperature is warm than the fall and in winter when soil is cool.

7. Sunlight:

Photodecomposition (Photolysis): Deactivation or decomposition of herbicide by sunlight. The UV band of light mostly induces photo decomposition, this may be result due to aromatic substitution, reduction elimination, cyclization and polymerization of herbicide molecule. Surface applied herbicides more prone to photo decomposition. Soil incorporation eliminate or reduce decomposition. E.g. In monuron photodecomposition in aqueous solution occurs through oxidation.

Absorption:

Herbicide absorption in roots in most cases is due to passive absorption and therefore factors affecting transpiration may affect absorption of herbicide by roots. Some major Environmental factors that affects the transpiration pull and translocation of herbicide along with water are discussed below:

Light:

Light, especially the light intensity is probably the most obvious among the Environmental factors affecting transpiration in plants. It has controlling effect on the opening of the stomata through which water primarily escapes in gaseous state. In general transpiration rate is higher during day time, particularly when light is bright, than in night.

Relative Humidity:

It affects transpiration by regulating stomatal movement and atmospheric demand. At high RH (moist air), the stomata tend to close and limit the exit of water vapour from the plant. Further high relative humidity means that the water potential gradient from plant to atmosphere will be minimal compared to when RH is low. In addition, at high RH the atmosphere contains

more water and low atmospheric demand, meaning that it has limited capacity to absorb more water. Transpiration rate will be faster at 50% RH, than at 90% RH.

If the humidity is more the uptake of herbicide is likely to be less due to decreased transpiration and reverse may be true if humidity is higher.

Temperature:

The rate of transpiration is fastest when air temperature is between 20-30° C (Moore *et al.*, 2013). In general stomata close at 0°C and progressively increases in aperture up to about 30°C (Delvin 1975). Increase in air temperature, decrease the plant water potential and thus enhance water translocation.

- If there increase in temperature greater uptake and translocation is expected to occur because of higher transpiration.
- Metabolism is also likely to take place at higher rate. Atrazine although selective may become phytotoxic to maize in wet and cool conditions due to excessive uptake, but not under hot and dry conditions.
- Under high temperature and moisture stress, the rate of penetration of herbicide molecule will be definitely reduced to a greater extent.

Wind:

It affects transpiration by removing thin moist layer of air, called boundary layer, which is replaced by drier air thus increasing water potential gradient and enhancing transpiration (Moore *et al.*, 2013). However strong wind may cause excessive loss of water from leaves leading to stomatal closure.

- In general plant transpire fastest under bright day, dry air, moist soil, warm temperature and windy day.

Rainfall:

Drizzling for a short period after herbicide application is useful to both soil and foliage applied herbicides. Little drizzling may incorporate surface applied herbicide into soil and increase their activity and selectivity. In dry soil (under rainfed conditions), light shower following herbicide application may increase the uptake.

However heavy rains may wash away the surface applied herbicides or there will be leaching of water soluble herbicides. The mechanism of selectivity of pre-emergence or pre-plant incorporated herbicides through depth protection may be affected since herbicide percolate to deeper layers of soil and may reach to the crop roots or seeds.

Research Findings

1. Experiment: Dinitroaniline herbicide phyto-toxicity as influenced by soil moisture and herbicide vaporization

Jacques and Harvey in 1979 performed laboratory experiments to study the effect of soil moisture and herbicide vapours on the phyto-toxicity of eight dinitroaniline herbicides *viz.* benefin, butralin, dinitramine, fluchloralin, oryzalin, profluralin, isopropalin and trifluralin to oat plant (*Avena sativa* L.).

- Oat primary root length was inhibited more by the herbicides than was shoot length or shoot fresh weight.
- All of these herbicides except oryzalin (3,5-dinitro-N, N-dipropylsulfanilamide) inhibited primary root length through water activity. Vapours of dinitramine were most inhibitory.
- Herbicide vapour inhibition increased with temperature.
- Soil water affected oryzalin activity more than it did that of another herbicide. Oryzalin Phyto-toxicity to the oat primary root was reduced more at low soil water than that of other herbicides. Its low vapour activity apparently reduced its effectiveness in dry soil.

2. Experiment: Persistence of pendimethalin and oxyfluorfen at different temperature and moisture levels in Alfisols and Vertisols.

Persistence of pendimethalin and oxyfluorfen in an alfisols and vertisols was studied at three moisture levels (at saturation, field capacity and 50% field capacity) and at two temperature levels (10 and 27°C) for 137 days in the laboratory. Degradation of pendimethalin and oxyfluorfen was more at higher temperature and moisture levels. The disappearance curve/semilogarithmic plot followed first order kinetics with two distinct pathways, an initial faster followed by slower and more gradual disappearance.

3. Experiment: Influence of organic matter, texture and availability of water on the toxicity of simazine in soil.

Addition of organic matter reduced the effectiveness of 2-chloro-4,6-bis(ethylamine)-s-triazine (simazine) to oats in heavy clay soil. At moisture levels, changes in the relative amounts of clay in the soil did not affect the toxicity of simazine. Toxicity of simazine decreased as the soil moisture level was reduced.

4. Experiment: Picloram degradation in soils as influenced by soil water content and temperature.

A laboratory experiment was conducted to determine the effect of temperature and alternating incubations at field capacity and during drying periods on the degradation of picloram (4-amino-3,5,6-trichloropicolinic acid) in five soils. Picloram was added at the rate of 10 ppm and degradation was measured by ¹⁴CO₂ evolution resulting from the cleavage of the labelled carboxyl carbon.

- Picloram degraded very little at 5°C and increased only slightly up to 25°C. Three soils were highest in degradation rate at 30°C while two soils were highest at 50°C. Picloram degradation rates during 20-day incubation period at field capacity interrupted with 16-day drying cycles, varied among soil and decreased after each successive drying cycle at 30 and 50°C except for one soil at 50°C.
- The degradation rate decreased gradually as water content decreased from field capacity to 15 bar tension and ceased after the soil were dried.

5. Experiment: Leaching behaviour of chlorothalonil, chlorpyrifos and pendimethalin in soil: Effect of soil organic matter and clay.

Leaching behaviour of chlorothalonil, chlorpyrifos and pendimethalin was studied in Inceptisol soil with normal, partially organic matter removed and clay fraction removed soil samples. In normal soil, the pesticide adsorbed strongly to soil surfaces leading to low leaching. About 84-87% of added pesticides were confined to the upper soil layer of depth 0-10cm. In partially organic matter removed soil (about 50%), about 2.5-6% of the pesticides were found in leachates compared to 2% in normal soil. Clay fraction controls the mobility behaviour of pesticides greatly. Most pronounced effect of clay on leaching was observed in chlorothalonil. Chlorothalonil leached out faster than other compounds in soils in which clay fraction was removed. The organic matter play important role in leaching behaviour of pesticides.

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INNOVATIVE STRATEGIES: NANO BIOFORMULATION IN TEA CROP PROTECTION

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Introduction:

Tea, *Camellia sinensis* (L.) O. Kuntze, has gained major praise as the world's most consumed beverage due to its flavour, aroma, and stimulating effect in addition to its medical or health benefits. Due to its diverse agroclimatic conditions, India is given to some of the best and most specialty teas in the world. The world enjoys Indian teas for their delicate flavours, brightness, and strength, especially those from Darjeeling, Assam, and the Nilgiris. Tea, a woody perennial with a long lifespan that grows in monoculture, creates an ideal microclimate and a steady source of food for the fast development of phytophagous species, which include numerous diseases, insects, and mites. According to Chen and Chen (1989), there are 1034 species of arthropods, 82 species of nematodes, an algal disease, and 350 fungal diseases linked to tea plants worldwide. A wide range of pest species, such as the tea mosquito bug, red spider mite, pink mites, thrips, termites, red slug caterpillar, looper, green leaf hopper, etc., infest tea plants in Northeastern India, severely reducing yields and degrading the quality of the tea leaves. Synthetic pesticides are used as a highly effective tool to control various insect pests and diseases in agricultural crops as a whole because of their broad host range action, although their detrimental effects on the environment and the overall sustainability of farming systems have received little attention. The over-reliance on synthetic chemicals for fertilisers and pesticides has resulted in a number of negative effects on agricultural food for humans and animals, as well as adverse effects on the environment's biotic and abiotic components.

There is an urgent need to investigate alternative pest control solutions due to the rising cost of insect pest management and growing concern about the harmful effects of pesticide residues in produced tea. Among many strategies, some satisfactory results have been reported using Nano technology as safe alternative to hazardous pesticides with negligible risk to human health and the environment.

Nano technology in enhancing crop productivity and sustainability in agriculture

Agrochemicals, which include fertilisers and insecticides, are essential to modern agriculture's ability to produce food efficiently and sustainably. But each agrochemical has certain possible drawbacks, such as the possibility for water contamination or residues on food products that threaten human and environmental health; therefore, careful input management and

control could help to lower these risks. Using engineered smart nanotools to develop a high-tech agricultural system could be a great way to change agricultural practices, lessen or even completely eradicate the adverse environmental impacts of modern agriculture, and increase yields in terms of both quality and quantity. Nanotechnology is an innovative, novel and scientific approach that leads to design, manipulation and development of useful nanomaterials. Nanotechnology generates materials in nanometer scale ranging in size from 1 to 100 nm (nm). Nano technology has the prospective to improve the agriculture with novel nanotools for the controlling of fast disease diagnostic, enhancing the capacity of plants to absorb nutrients among others. Utilising nanotechnology in agriculture has many benefits, including the development of targeted applications like nano pesticides and nano fertilizers that raise productivity levels without contaminating soils or waterways and provide protection against a variety of microbial diseases and insect pests.

Nano Bioformulation: An alternative technology for managing insect pest and diseases in tea

Nano bioformulation involves the integration of nanotechnology with biological agents to create potent crop protection solutions. At the nanoscale, materials exhibit unique properties that can enhance efficacy, stability, and targeted delivery of bioactive compounds. In the context of tea crop protection, nano bioformulation offers a promising avenue for combatting pests and diseases with precision and efficiency. Due to their excellent stability and biodegradability, nanoparticles have found success in the manufacturing of nanocapsules, which are used to deliver agrochemicals such as fertilisers and pesticides. As insecticides, nanomaterials containing polymeric, silver, gold, and iron oxide nanoparticles are employed.

The term "nanopesticides" refers to a formulation that purposefully contains substances in the nm size range or makes claims about unique capabilities connected to these small size range. According to reports, nanoparticles in insects have the potential to be used as pesticides (Bhattacharyya *et al.*, 2010). Nano materials can deliver biocontrol agents or beneficial compounds precisely to tea plants, enhancing their resistance to pests and diseases.

Now a days the Biological Control Agents (BCAs), such as Beauveria, Metarhizium, and other species, have been shown to be safe and promising components of IPM and organic cultivation strategies used in several crops, including tea. Microorganisms are the most important and active part of many agroecosystems, including the tea ecosystem. They can be found in soil, air, and phylloplane, and they have a variety of known and unknown interactions with plants that greatly influence crop output. It was reported that 72 viral species and approximately 40 fungal and bacterial species that are efficient against tea insect and mite pests have been found as possible biocontrol agents for tea plants, and other viruses and fungi have been studied to identify others. Different isolations of *B. bassiana* have been shown to be effective against the tea mosquito bug as well as other insect pests of tea plants such as tea weevils and termites. The efficacy of *B. bassiana* in both laboratory and field circumstances may be attributed to the development of various toxins during infection, including beauvericin, enniatins, oosporein, and bassianolide, which play an important role in *B. bassiana's* pathogenic

activity against the tea mosquito bug. On the other hand, using *M. anisopliae* instead of a traditional termiticide resulted in a considerable reduction in termite infestation. *M. anisopliae* has previously been shown to be effective against mite pests of several crops, including tea, such as tea red spider mite. *Bacillus thuringiensis* is a sporogenous gram-positive bacterium found all over the world which is widely used to control tea pests, and it has been shown to be 95% effective against lepidopterous larvae.

Nano bioformulation: A multi-faceted approach

- 1. Targeted Delivery System:** Nano sized carriers enable precise delivery of bioactive compounds to specific sites within the tea plant, maximizing efficiency while minimizing environmental impact.
- 2. Enhanced Bioavailability:** Nano formulations enhance the solubility and absorption of active ingredients, ensuring optimal utilization by tea plants for robust defense mechanisms.
- 3. Longer Residual Activity:** Nano encapsulation prolongs the release and activity of bioactive compounds, providing sustained protection against pests and diseases over extended periods.
- 4. Reduced Environmental Footprint:** By minimizing the use of synthetic pesticides and fertilizers, nano bioformulation contributes to sustainable tea cultivation practices, promoting environmental stewardship.

Challenges and Future Directions:

Despite the immense potential of nano bioformulation, several challenges remain, including regulatory hurdles, scalability issues, and potential ecological impacts. Future research efforts should focus on addressing these challenges while further optimizing nano formulations for commercial adoption in tea crop protection. Collaboration between academia, industry, and regulatory agencies is essential to accelerate the translation of innovative nano technologies from the laboratory to the field.

Conclusion:

Innovative strategies such as nano bioformulation hold great promise for revolutionizing tea crop protection. By harnessing the unique properties of nanomaterials and biological agents, nano formulations offer a sustainable and effective approach to combatting pests and diseases while minimizing environmental impact. Continued research and investment in this field are crucial to realizing the full potential of nano bioformulation in ensuring the long-term sustainability and resilience of tea agriculture worldwide.

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MARKER ASSISTED SELECTION IN CROP BREEDING

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Abstract:

Genetic mapping of major genes and quantitative traits loci (QTLs) for many important agricultural traits is increasing the integration of biotechnology with the conventional breeding process. Exploitation of the information derived from the map position of traits with agronomical importance and of the linked molecular markers, can be achieved through marker assisted selection (MAS) of the traits during the breeding process. However, empirical applications of this procedure have shown that the success of MAS depends upon several factors, including the genetic base of the trait, the degree of the association between the molecular marker and the target gene, the number of individuals that can be analyzed and the genetic background in which the target gene has to be transferred. MAS for simply inherited traits is gaining increasing importance in breeding programs, allowing an acceleration of the breeding process. For improving polygenic traits in a quick time-frame and in a cost effective manner, recent advances in MAS strategies have suggested improved selection schemes in which MAS is applied only once during the breeding process to arrange of agronomically important traits or involve the mapping of loci for all agronomic important traits and selection schemes in which MAS operates by cyclically re-estimating the value of the QTL alleles each time a new set of germplasm is generated during the breeding process. Traits related to disease resistance to pathogens and to the quality of some crop products are offering some important examples of a possible routinary application of MAS. For more complex traits, like yield and abiotic stress tolerance, a number of constraints have determined severe limitations on an efficient utilization of MAS in plant breeding, even if there are a few successful applications in improving quantitative traits. Recent advances in genotyping technologies together with comparative and functional genomic approaches are providing useful tools for the selection of genotypes with superior agronomical performances.

Keywords: Crop Improvement, Genetic Mapping, Genome Analysis, Marker Assisted Selection, PCR-Based Markers, QTLs, Syteny.

Marker Assisted Selection in Crop Breeding

Introduction:

Conventional plant breeding is primarily based on phenotypic selection of superior individuals among segregating progenies resulting from hybridization. Although significant strides have been made in crop improvement through phenotypic selections for agronomically

important traits, considerable difficulties are often encountered during this process, primarily due to genotype – environment interactions. Besides, testing procedures may be many times difficult, unreliable or expensive due to the nature of the target traits (e.g. abiotic stresses) or the target environment.

Marker-Assisted Selection (MAS) is a powerful tool in crop breeding that enables plant breeders to select plants with desired traits more efficiently and accurately. It involves the use of molecular markers, such as DNA sequences or proteins, to identify and select plants with specific genes or traits of interest. This method has revolutionized crop breeding by accelerating the breeding process and increasing the precision of trait selection. Here's how marker-assisted selection works in crop breeding:

- **Trait Identification:** The first step in MAS is to identify the target trait or gene of interest. This could be a specific disease resistance gene, a gene for high yield, or any other desirable trait.
- **Marker Development:** Once the target trait or gene is identified, molecular markers are developed that are closely linked to the trait. Common molecular markers include single nucleotide polymorphisms (SNPs), microsatellites, or amplified fragment length polymorphisms (AFLPs). These markers are specific sequences of DNA that can be easily detected and genotyped.
- **Genotyping:** In the genotyping step, DNA samples from a population of plants are analyzed to determine the presence or absence of the marker sequences. This is typically done through techniques such as polymerase chain reaction (PCR) or sequencing.
- **Data Analysis:** The genotyping data is then analyzed to determine which plants carry the desired trait or gene based on the presence or absence of the molecular markers. This data helps breeders make informed decisions about which plants to select for further breeding.
- **Selection of Plants:** Using the marker information, breeders can select plants with the target trait or gene with high accuracy. This is a significant improvement over traditional breeding methods, which rely on visual assessments and may require several generations to achieve the desired trait.
- **Crossover and Backcrossing:** Selected plants can be crossed with other plants to introduce additional genetic diversity. Backcrossing can be used to gradually introduce the trait into the background of elite or commercial varieties, which can make the new variety more acceptable to growers and consumers.
- **Field Testing:** The selected plants and their offspring are grown and evaluated in field trials to ensure that they perform well under real-world conditions.
- **Commercialization:** Once a new variety is deemed successful in field trials, it can be released for commercial production.

Advantages of Marker-Assisted Selection in Crop Breeding:

- **Increased Precision:** MAS allows for the selection of plants with the target trait at a very early stage, increasing the precision of breeding efforts.
- **Faster Breeding:** It accelerates the breeding process by reducing the time required to develop new crop varieties.
- **Reduced Costs:** It can reduce the cost of field trials, as plants are pre-selected based on genetic markers before field testing.
- **Disease Resistance:** It is particularly valuable for identifying and incorporating disease-resistant genes into crop varieties.
- **Preservation of Desired Genetic Background:** MAS allows breeders to preserve the genetic background of elite varieties while introducing specific traits, which is critical for maintaining desirable agronomic characteristics. Marker-assisted selection has significantly advanced the field of crop breeding, enabling the development of new varieties with improved traits and resistance to diseases and pests. It has the potential to contribute to global food security by increasing the efficiency and precision of crop improvement.

Molecular marker-assisted selection, often simply referred to as marker-assisted selection (MAS) involves selection of plants carrying genomic regions that are involved in the expression of traits of interest through molecular markers. With the development and availability of an array of molecular markers and dense molecular genetic maps in crop plants, MAS has become possible for traits both governed by major genes as well as quantitative trait loci (QTLs). A generalized and simplified key for distinction between qualitative and quantitative traits¹ is presented in Table 1. If the individual genes or QTLs with significant influence on specific target trait(s) can be identified based on their linkage to molecular markers, the efficiency of incorporating the desired traits in elite germplasm could be greatly enhanced. While plenty of information is already available on different kinds of marker systems and gene–marker associations, the practicalities of designing an MAS strategy, increasing their chances of success and efficiency have not received adequate attention till now.

Salient requirements for MAS: In general, the success of a marker-based breeding system depends on these main factors:

- (i) A genetic map with an adequate number of uniformly-spaced polymorphic markers to accurately locate desired QTLs or major genes.
- (ii) Close linkage between the QTL or a major gene of interest and adjacent markers.
- (iii) Adequate recombination between the markers and rest of the genome.
- (iv) An ability to analyse a larger number of plants in a time and cost-effective manner.

The success of MAS depends on location of the markers with respect to genes of interest. Three kinds of relationships between the markers and respective genes could be distinguished. The success of MAS depends on location of the markers with respect to genes of

interest. Three kinds of relationships between the markers and respective genes could be distinguished.

Strategies for Marker-Assisted Selection: The recent advances in genomics have paved the way for clear and reliable methods for MAS in plants: from QTL identification, NIL development and fine-mapping to transferring the QTL into popular varieties using a precise marker-assisted backcrossing (MABC) strategy (Mackill 2006, Collard *et al.*, 2005, Collard, Mackill 2008 and Collard *et al.*, 2008). MABC involves the manipulation of genomic regions involved in the expression of particular traits of interest through DNA markers, and combines the power of a conventional backcrossing program with the ability to differentiate parental chromosomal segments. The efficiency of a MABC program depends on a number of factors, including the size and reliability of the target QTL effect, the precision of the target gene/QTL fine-map, the rate of polymorphism when identifying background markers, as well as the cost, speed, and failure rate of the markers employed in each customized MABC system. For each set of parents and for each target QTL, a customized MABC package needs to be developed with optimized foreground markers to select the QTL target, recombinant markers flanking the locus of interest to reduce linkage drag and an evenly-spaced set of polymorphic background markers across the genome to select the recurrent parent background (Neeraja *et al.*, 2007, Collard and Mackill 2008).

Foreground Selection and Background Selection: Molecular markers are now increasingly being employed to trace the presence of target genes (foreground selection) as well as for accelerating the recovery of the recurrent parent genome (background selection) in backcross programmes.

Marker Assisted Selection in Crop Breeding: Backcrossing has been a widely used technique in plant breeding for almost a century. Backcrossing is a plant breeding method most commonly used to incorporate one or a few genes into an adapted or elite variety. In most cases, the parent used for backcrossing has a large number of desirable attributes but is deficient in only a few characteristics (Allard 1999). The method was first described in 1922 and was widely used between the 1930s and 1960s (Stoskopf *et al.*, 1993).

Marker-Assisted Backcrossing (MAB) improves the efficiency of backcross breeding in three ways:

- (i) If the phenotype of the desired gene cannot be easily assayed, backcross (BC) progeny possessing a marker allele from the donor parent at a locus near/within the target gene can be selected with a good probability of carrying the gene
- (ii) Markers can be used to select BC progeny with least amounts of donor parent germplasm in the genome outside the target region
- (iii) Markers can be used to select rare progeny that are the result of recombination near the target gene, thus minimizing the effects of linkage drag.

Improving Qualitative Traits using MAS:

(a) **MAS for resistance to soybean cyst nematode:** Disease resistant phenotypes are often simple and oligogenic in nature. Yet, the difficulties in establishing reliable inoculation and scoring methods can challenge even the best plant pathologist or breeder. An example is the resistance to soybean cyst nematode (*Heterodera glycines*). MAS in developing QPM genotypes: Normal maize protein is inherently deficient in two essential amino acids, namely lysine and tryptophan. QPM refers to maize genotypes in which the opaque2 gene is introgressed along with endosperm modifiers that provide the hard kernel texture.

(b) **Marker-aided pyramiding of rice genes for bacterial blight and blast resistance:** The most effective approach to combat bacterial blight caused by *Xanthomonas oryzae* pv. *oryzae* is the use of resistant varieties. So far, 25 resistance genes have been identified¹⁶ and some of them have been incorporated into modern rice varieties through conventional breeding.

MAS for improvement of quantitative traits

Most of the traits of agronomic importance are complex and regulated by several genes. Unlike the case of simply inherited traits that are controlled by one or a few major genes, improvement of polygenic traits through MAS is a complex endeavor. The difficulty in manipulating quantitative traits is related to their genetic complexity, mainly the number of genes involved in their expression and interactions among genes (epistasis).

Despite the proliferation of QTL mapping experiments in recent years, a number of constraints have imposed severe limitations on efficient utilization of QTL mapping information in plant breeding through Example of the marker-assisted backcrossing scheme used to transfer the *SUB1* QTL for submergence tolerance into six mega-varieties, showing recommended numbers of plants and markers for each step to develop a Sub1-converted near-isogenic line (NIL), at either the BC2F2 or BC3F2 generation, depending on the number of background introgressions remaining at the BC2F1 generation and the size of the target introgression desired (Neeraja *et al.*, 2007).

Constraints in MAS:

- (a) Identification of a limited number of major (QTLs) controlling specific traits.
- (b) The notion that QTL identification is required whenever additional germplasm is used.
- (c) Inadequacies/ experimental deficiencies in QTL analysis leading to either overestimation or underestimation of the number and effects of QTLs.
- (d) Lack of universally valid QTL marker associations applicable over different sets of breeding materials.
- (e) Strong QTL – environment interaction.
- (f) Difficulty in precisely evaluating epistatic effects.

Quantitative Trait Improvement through MAS:

(a) **MAS for improving heterotic performance in maize:** The maize lines B73 and Mo17 are not highly productive, but widely used across the world, and represent the two most important ‘heterotic groups’ in the US maize breeding programmes. Based on a mapping population

derived from the B73 × Mo17 cross, QTLs contributing to heterosis for grain yield were mapped on nine of the ten maize chromosomes. Further, mapping studies suggested that two elite inbred lines, Tx303 and Oh43 contained genetic factors that might improve the heterotic response of the B73 × Mo17 single cross hybrid.

(b) MAS for drought stress tolerance in maize: Researchers have made considerable efforts during the past three decades to improve pre and post flowering drought tolerance in maize. Although significant progress has been achieved for improving drought tolerance in CIMMYT maize germplasm through conventional breeding, the approach is slow and time-consuming. Use of molecular markers and QTL information based on carefully managed replicated tests showed the potential to alleviate the problems associated with inconsistent and unpredictable onset of moisture stress or the confounding effect of other stresses such as heat.

Examples of marker-assisted backcrossing in cereals

Species	Traits	Gene/QTLs	Foreground selection	Background selection	Reference
Barley	Barley yellow Dwarf virus	<i>Yd2</i>	STS	not performed	Jefferies <i>et al.</i> (2003)
Barley	Leaf rust	<i>Rphq6</i>	AFLP	AFLP	Van Berloo <i>et al.</i> (2001)
Barley	Stripe rust	QTLs on 4Hand 5H	RFLP	not performed	Toojinda <i>et al.</i> (1998)
Barley	Yield	QTLs on 2HL and 3HL	RFLP	RFLP	Schmierer <i>et al.</i> (2004)
Maize	Corn borer Resistance	QTLs on chromosomes 7,9 and 10	RFLP	RFLP	Willcox <i>et al.</i> (2002)
Maize	Earliness and yield	QTLs on chromosomes 5, 8 and 10	RFLP	RFLP	Bouchez <i>et al.</i> (2002)
Rice	Bacterial blight	<i>Xa21</i>	STS	RFLP	Chen <i>et al.</i> (2000)

Recent Advances in MAS Strategies and Genotyping Techniques:

Single large-scale MAS: In this strategy, selection of parental lines is first carried out among outstanding elite material for the trait to be improved with the best allelic complementarity. By crossing each selected parental line with a tester (elite line lacking the target trait), segregating populations are developed. Genomic regions of interest for each parental line are identified by combining favourable alleles in the segregating populations (e.g. F3 families and RILs). MAS, based on reliable PCR-based markers to fix favourable alleles at target

genomic regions, is conducted only once on large segregating populations derived from crosses between the elite lines.

Pedigree MAS: This approach is especially relevant for crops such as wheat, where pedigrees of elite germplasm are known. Fingerprinting elite wheat materials must be conducted in a set of lines actively used in the breeding programme, and in elite materials to be subsequently released. The data may be combined with the phenotypic data collected during different selection cycles to identify favourable alleles for traits of interest. For example, if an elite line contains alleles for yield performance in a target environment, their frequency should be higher than the expected random frequency in offspring derived from this elite parental line.

Breeding by Design: 'Breeding by design' is a novel concept that aims to control all allelic variations for all genes of agronomic importance. The authors of this concept⁴² propose that by understanding the genetic basis of all agronomically important characters and the allelic variation at those loci, the breeder would be able to design superior genotypes *in silico*, which demonstrates that DNA markers not only improve selection processes but can aid in creating novel genotypes bearing new characteristics of agronomic importance.

The strategy involves well-defined steps:

- (a) Mapping loci involved in all agronomically relevant traits, preferably through introgression line (IL) libraries that are extremely powerful not only in mapping, but also in reducing the complexity of polygenic traits by separating them into a set of monogenic loci. An IL library consists of a series of lines harboring a single homozygous donor segment introgressed into a uniform cultivated background.
- (b) Assessment of the allelic variation at those loci through intensive chromosome haplotyping and extensive phenotyping of all agronomic traits. Haplotypes refer to a combination of linked marker alleles that occur in a locus in a set of accessions.
- (c) Designing superior genotypes comprising a combination of favourable alleles at all loci through accurate selection of recombination events using flanking markers to collate different favourable alleles next to each other. 'Breeding by design' involves an integrative, complementary application of technological tools and materials currently available to develop superior varieties. However, success of this approach would essentially depend upon availability of extremely saturated marker maps and precision of phenotyping.

Current Status of Applications of MAS in Agriculture:

Most of the traits of agronomic importance are complex and regulated by several genes. Unlike the case of simply inherited traits that are controlled by one or a few major genes, improvement of polygenic traits through MAS is a complex endeavour. The difficulty in manipulating quantitative traits is related to their genetic complexity, mainly the number of genes involved in their expression and interactions among genes (epistasis).

Applications of MAS in Plant Breeding

The advantages described above may have a profound impact on plant breeding in the future and may alter the plant breeding paradigm (Koeber and Summers 2003). In this section, we

describe the main uses of DNA markers in plant breeding, with an emphasis on important MAS schemes. We have classified these schemes into four broad areas: marker-assisted evaluation of breeding material, pyramiding and combined MAS, although there may be overlap between these categories. Generally, for line development, DNA markers have been integrated in conventional schemes or used to substitute for conventional phenotypic selection.

a) Marker-Assisted Evaluation of Breeding Material

Prior to crossing (hybridization) and line development, there are several applications in which DNA marker data may be useful for breeding, such as cultivar identity, assessment of genetic diversity and parent selection, and confirmation of hybrids. Traditionally, these tasks have been done based on visual selection and analysing data based on morphological characteristics.

(i) Cultivar Identity/Assessment of ‘Purity’

In practice, seed of different strains is often mixed due to the difficulties of handling large numbers of seed samples used within and between crop breeding programmes. Markers can be used to confirm the true identity of individual plants. The maintenance of high levels of genetic purity is essential in cereal hybrid production in order to exploit heterosis. In hybrid rice, SSR and STS markers were used to confirm purity, which was considerably simpler than the standard ‘grow-out tests’ that involve growing the plant to maturity and assessing morphological and floral characteristics (Yashitola *et al.*, 2002).

(ii) Assessment of Genetic Diversity And Parental Selection

Breeding programmes depend on a high level of genetic diversity for achieving progress from selection. Broadening the genetic base of core breeding material requires the identification of diverse strains for hybridization with elite cultivars (Xu *et al.*, 2004 and Reif *et al.*, 2005). Numerous studies investigating the assessment of genetic diversity within breeding material for practically all crops have been reported. DNA markers have been an indispensable tool for characterizing genetic resources and providing breeders with more detailed information to assist in selecting parents. In some cases, information regarding a specific locus (e.g. a specific resistance gene or QTL) within breeding material is highly desirable. For example, the comparison of marker haplotypes has enabled different sources of resistance to *Fusarium* headblight, which is a major disease of wheat worldwide, to be predicted (Liu & Anderson 2003 and McCartney *et al.*, 2004).

(iii) Study of Heterosis:

For hybrid crop production, especially in maize and sorghum, DNA markers have been used to define heterotic groups that can be used to exploit heterosis (hybrid vigour). The development of inbred lines for use in producing superior hybrids is a very time-consuming and expensive procedure. Unfortunately, it is not yet possible to predict the exact level of heterosis based on DNA marker data although there have been reports of assigning parental lines to the proper heterotic groups (Lee *et al.*, 1989 and Reif *et al.*, 2003). The potential of using smaller subsets of DNA marker data in combination with phenotypic data to select heterotic hybrids has also been proposed (Jordan *et al.*, 2003).

(iv) Identification of Genomic Regions Under Selection

The identification of shifts in allele frequencies within the genome can be important information for breeders since it alerts them to monitor specific alleles or haplotypes and can be used to design appropriate breeding strategies (Steele *et al.*, 2004). Other applications of the identification of genomic regions under selection are for QTL mapping: the regions under selection can be targeted for QTL analysis or used to validate previously detected marker–trait associations (Jordan *et al.*, 2004). Ultimately, data on genomic regions under selection can be used for the development of new varieties with specific allele combinations using MAS schemes such as marker-assisted backcrossing or early generation selection (Ribaut *et al.*, 2001 and Steele *et al.*, 2004).

(b) Marker-Assisted Pyramiding

Pyramiding is the process of combining several genes together into a single genotype. Pyramiding may be possible through conventional breeding but it is usually not easy to identify the plants containing more than one gene. Using conventional phenotypic selection, individual plants must be evaluated for all traits tested. Therefore, it may be very difficult to assess plants from certain population types or for traits with destructive bioassays. DNA markers can greatly facilitate selection because DNA marker assays are non-destructive and markers for multiple specific genes can be tested using a single DNA sample without phenotyping.

The most widespread application for pyramiding has been for combining multiple disease resistance genes (i.e. combining qualitative resistance genes together into a single genotype). Some evidence suggests that the combination of multiple genes (effective against specific races of a pathogen) can provide durable (broad spectrum) resistance (Kloppers & Pretorius 1997, Shanti *et al.*, 2001 and Singh *et al.*, 2001). The ability of a pathogen to overcome two or more effective genes by mutation is considered much lower compared with the ‘conquering’ of resistance controlled by a single gene. In the past, it has been difficult to pyramid multiple resistance genes because they generally show the same phenotype, necessitating a progeny test to determine which plants possess more than one gene. With linked DNA markers, the number of resistance genes in any plant can be easily determined. The incorporation of quantitative resistance controlled by QTLs offers another promising strategy to develop durable disease resistance. Castro *et al.*, (2003) referred to quantitative resistance as an insurance policy in case of the breakdown of qualitative resistance.

Pyramiding may involve combining genes from more than two parents. For example, Hittalmani *et al.*, (2000) and Castro *et al.*, (2003) combined genes originating from three parents for rice blast and stripe rust in barley, respectively. MAS pyramiding were also proposed as an effective approach to produce three-way F1 cereal hybrids with durable resistance (Witcombe and Hash 2000). Strategies for MAS pyramiding of linked target genes have also been evaluated (Servin *et al.*, 2004). For many linked target loci, pyramiding over successive generations is preferable in terms of minimizing marker genotyping.

(c) Early Generation Marker-Assisted Selection

Although markers can be used at any stage during a typical plant breeding programme, MAS is a great advantage in early generations because plants with undesirable gene combinations can be eliminated. This allows breeders to focus attention on a lesser number of high-priority lines in subsequent generations. When the linkage between the marker and the selected QTL is not very tight, the greatest efficiency of MAS is in early generations due to the increasing probability of recombination between the marker and QTL. The major disadvantage of applying MAS at early generations is the cost of genotyping a larger number of plants.

The population sizes may soon become quite small due to the high selection pressure, thus providing an opportunity for genetic drift to occur at non-target loci, so it is recommended that large population sizes be used (Ribaut and Betran 1999). This problem can also be minimized by using F3 rather than F2 populations, because the selected proportion of an F3 population is larger compared with that of an F2 population (i.e., for a single target locus, 38% of the F3 population will be selected compared with 25% of the F2). Ribaut & Betran (1999) also proposed that, theoretically, linkage drag could be minimized by using additional flanking markers surrounding the target QTLs, much in the same way as in MAB.

D) Combined Marker-Assisted Selection

There are several instances when phenotypic screening can be strategically combined with MAS. In the first instance, 'combined MAS' (coined by Moreau *et al.*, 2004) may have advantages over phenotypic screening or MAS alone in order to maximize genetic gain (Lande and Thompson 1990). This approach could be adopted when additional QTLs controlling a trait remain unidentified or when a large number of QTLs need to be manipulated. Simulation studies indicate that this approach is more efficient than phenotypic screening alone, especially when large population sizes are used and trait heritability is low (Hospital *et al.*, 1997). Bohn *et al.*, (2001) investigated the prospect of MAS for improving insect resistance in tropical maize and found that MAS alone was less efficient than conventional phenotypic selection. However, there was a slight increase in relative efficiency when MAS and phenotypic screening were combined. In an example in wheat, MAS combined with phenotypic screening was more effective than phenotypic screening alone for a major QTL on chromosome 3BS for Fusarium head blight resistance (Zhou *et al.*, 2003). In practice, all MAS schemes will be used in the context of the overall breeding programme and this will involve phenotypic selection at various stages. This will be necessary to confirm the results of MAS as well as select for traits or genes for which the map location is unknown.

Plant breeding in the future: The dawn of marker-assisted selection?

Despite the relatively small impact that MAS has had on variety development to date, there has been a 'cautious optimism' for the future (Young 1999). We predict that six main factors will give rise to a much greater level of adoption of MAS in plant breeding in the early part of the twenty-first century in many breeding programs.

First, the extent to which DNA marker technology has already spread to plant breeding institutes coupled with the enormous amount of data from previous QTL mapping and MAS studies should lead to the greater adoption of MAS. Many such institutes now possess the essential equipment and expertise required for marker genotyping. Of course, the frequency of use will depend on available funding.

Second, since the landmark concept of 'advanced BC QTL analysis' directly integrated QTL mapping with plant breeding by combining QTL mapping with simultaneous variety development (Tanksley and Nelson 1996), there have been several encouraging examples of an efficient merging of plant and molecular breeding. Some of these excellent examples are Toojinda *et al.*, (1998) and Castro *et al.*, (2003) in which QTL mapping and MAS breeding were combined. There have also been encouraging reports of the combination of QTL validation and line development (Flint-Garcia *et al.*, 2003). The use of backcrossing and the development of near-isogenic lines (NILs) may be particularly advantageous in this context (Stuber *et al.*, 1999 and van Berloo *et al.*, 2001). Ideally, QTL mapping and marker-assisted line development should now always be conceived together, in a holistic scheme.

Third, the increasing use of genetic transformation technology means that MAS can be used to directly select for progeny that possess transgenes via target gene selection. As discussed earlier, specific genotypes often with poor agronomic characteristics are routinely used for transformation. Therefore, MAS can be used to track the transgenes during elite line development.

Fourth, a rapid growth in genomics research has taken place within the last decade. Data generated from functional genomics studies have led to the identification of many candidate genes for numerous traits. SNPs within candidate genes could be extremely useful for 'association mapping' and ultimately MAS (Rafalski 2002, Flint-Garcia *et al.*, 2003, Gupta *et al.*, 2005, Breseghello and Sorrells 2006). This approach also circumvents the requirement for constructing linkage maps and performing QTL analysis for new genotypes that have not been previously mapped, although genotyping and phenotyping of segregating populations (e.g. F2 or F3) is recommended for marker validation (Breseghello and Sorrells 2006). Furthermore, genome sequencing projects in rice and other crop species will provide considerable data that could be used for QTL mapping and marker development in other cereals (Gale & Devos 1998, Yuan *et al.*, 2001 and Varshney *et al.*, 2005). However, the costs associated with genomics may be considerable. This could be detrimental to breeding programmes if funding is diverted away from actual breeding efforts (Brummer 2004).

Fifth, many new high-throughput methods for DNA extraction and especially new high-throughput marker genotyping platforms have been developed (Syvanen 2001, 2005). A current trend in some crops is the adoption of high-throughput genotyping equipment for SSR and SNP markers, although the cost of these new platforms may be higher than for standard genotyping methods (Brennan *et al.*, 2005). Some of these genotyping platforms use fluorescently labelled primers that permit high levels of multiplexing (Coburn *et al.*, 2002). Some authors have

predicted that SNP markers, due to their widespread abundance and potentially high levels of polymorphism, and the development of SNP genotyping platforms will have a great impact on MAS in the future (Rafalski 2002, Koebner and Summers 2003). Numerous SNP genotyping platforms have been recently developed, usually for medical applications; however, at present no superior platform has been universally adopted (Syvanen 2001). Array-based methods such as Diversity Array Technology DArT (Jaccoud *et al.*, 2001) and single feature polymorphism (SFP) detection (Hazen and Kay 2003 and Rostoks *et al.*, 2005) offer prospects for lower-cost marker technology that can be used for whole-genome scans.

Finally, the availability of large numbers of publicly available markers and the parallel development of user-friendly databases for the storage of marker and QTL data will undoubtedly encourage the more widespread use of MAS. In cereals, two of the most extensive and useful databases are 'Gramene' and 'Grain Genes' (Ware *et al.*, 2002 and Matthews *et al.*, 2003). The development and curation of these and other databases to keep pace with the continually growing amount of data generated will be critical for the efficient use of markers in the future (Lehmensiek *et al.*, 2005).

Although we believe that these factors will lead to the greater adoption of MAS in many instances (especially for major cereals), there will clearly be situations in which the incorporation of MAS in plant breeding programmes will still be very slow or even non-existent, for example in orphan crop species and in developing countries (Naylor *et al.*, 2004). In both of these situations, funding of research and breeding programmes is extremely limited. The improvement of orphan crop species, especially in developing countries—using any method—represents another great challenge for agricultural scientists.

Generally, the cost of MAS will continue to be a major obstacle for its application. It should be noted that MAS cost estimates may change depending on the number of samples and/or number of marker assays. The study by Dreher *et al.*, 2003 indicated that costs may decrease as the number of samples and/or marker assays increases due to economies of scale and lack of divisibility for many components of MAS. One current trend is the establishment of marker genotyping companies, which will enable marker genotyping to be outsourced. Assuming that the costs for outsourcing genotyping are cheaper, and that logistical problems are not created or are minimal, this may provide breeding programmes with more opportunities for MAS. Furthermore, some new SNP high-throughput genotyping methods may also be comparable with or even cheaper than current methods, although a large initial investment is required for the purchase of equipment (Chen and Sullivan 2003).

Conclusion:

Plant breeding has made remarkable progress in crop improvement and it is critical that this continue. It seems clear that current breeding programmes continue to make progress through commonly used breeding approaches. MAS could greatly assist plant breeders in reaching this goal although, to date, the impact on variety development has been minimal. For the potential of MAS to be realized, it is imperative that there should be a greater integration

with breeding programmes and that current barriers be well understood and appropriate solutions developed. The exploitation of the advantages of MAS relative to conventional breeding could have a great impact on crop improvement. The high cost of MAS will continue to be a major obstacle for its adoption for some crop species and plant breeding in developing countries in the near future. Specific MAS strategies may need to be tailored to specific crops, traits and available budgets. New marker technology can potentially reduce the cost of MAS considerably. If the effectiveness of the new methods is validated and the equipment can be easily obtained, this should allow MAS to become more widely applicable for crop breeding programmes.

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BIOAVAILABILITY, TOXICITY AND FATE OF NANOMATERIALS IN AGROECOSYSTEMS

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Abstract:

Nanotechnology holds immense promise in revolutionizing agriculture through improved crop yields, nutrient delivery, and pest management. However, the introduction of nanomaterials into agroecosystems raises concerns regarding their potential adverse effects on the environment, human health, and biodiversity. This book chapter provides a comprehensive analysis of the bioavailability, toxicity, and fate of nanomaterials in agroecosystems. The mechanisms governing the interaction between nanomaterials and various components of agroecosystems, includes soil, water, plants, and microorganisms. Understanding these interactions is crucial for predicting the behavior of nanomaterials in the environment and assessing their potential risks. Next, we delve into the factors influencing the bioavailability of nanomaterials in agroecosystems, such as particle size, surface chemistry, and environmental conditions. The uptake of nanomaterials by plants and their subsequent translocation within the plant are examined, along with the implications for food safety and human health. Furthermore, we discuss the potential toxicity of nanomaterials to agroecosystem organisms, including plants, soil microorganisms, and beneficial insects. We explore the mechanisms underlying nanotoxicity and highlight the importance of standardized toxicity testing protocols for assessing the safety of nanomaterials in agriculture. Finally, we address the fate and persistence of nanomaterials in agroecosystems, including their degradation pathways, accumulation in soil and water, and long-term environmental impacts. Strategies for mitigating the risks associated with nanomaterials in agriculture, such as green synthesis approaches and eco-friendly disposal methods, are also discussed. Overall, this book chapter provides valuable insights into the complex interactions between nanomaterials and agroecosystems, emphasizing the importance of holistic risk assessment and sustainable nanotechnology practices in ensuring the safe and responsible deployment of nanomaterials in agriculture.

Introduction:

With limited water and land resources, targeted agricultural growth can be obtained by increasing per unit natural resources productivity and farm income through judicious use of

advanced technologies. Using short-duration improved high-yielding varieties, intense fertiliser use, irrigation, and pesticides, potential yields and farm earnings were increased manyfold during the green revolution era, neglecting sustainable and effective use of natural resources.

Recently, farm productivity and incomes have shown steady decline. Since 60% of Indians rely on agriculture for their living, stabilising agricultural production is necessary to sustain the country's overall growth. The problems are accelerated by deterioration of natural resources like soil, climate, and water. This difficult situation of Indian agriculture is known as “technology fatigue.” To overcome the “technology fatigue,” focusing on technologies that can increase agricultural production, resource use efficiencies, and product quality is the need of the hour. Among the many scientific advancements, nanotechnology (NT) has been identified as one of the potential technologies that can revive the agriculture and food industry (Kuzma and VerHage 2006) and can improve livelihood of poor people.

The term “nanotechnology” was first used by Norio Taniguchi in 1974. Nanotechnology is the branch of science which studies the understanding of matter at nanometer dimensions (1–100 nm (US EPA). It manipulates matter of individual atoms, molecules, and molecular clusters in new structures with new or entirely different properties. It involves varied applications like building, controlling, and structuring of devices and materials of various nature like chemical, physical, and biological at nanoscale level.

However, at global level as well as in India, application of nanotechnology in agricultural sector and food systems is at nascent stage, and its ultimate success will depend on the acceptance of the stakeholders. Application of nanotechnology in agriculture calls for an effective regulatory and strong governance mechanisms and system with all group of stakeholders’ involvement. Thus, nanotechnology can provide the much required second green revolution in Indian agricultural sector with emphasis on sustainable production.

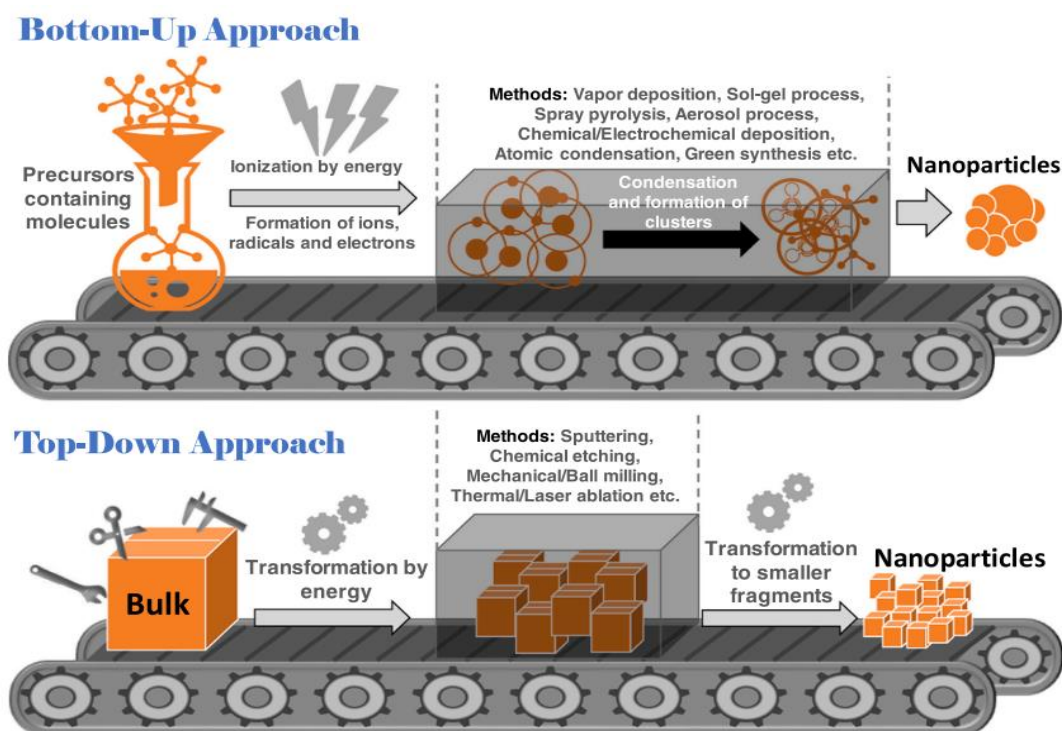
In recent years, new methods of product synthesis and development of nano-sized materials has quickly advanced globally. Due to such widescale use, nanomaterials are expected to eventually reach the environment due to their release from normal wear on mechanical systems, industrial and consumer products, and waste sources. In particular, it is anticipated that the majority of nanomaterials entering wastewater streams will eventually be partitioned to sewage sludge (Limbach *et al.*, 2008; Luoma, 2008; Mueller and Nowack 2008; Kiser *et al.*, 2009), where a large proportion is ultimately applied to land as biosolids. Therefore, terrestrial ecosystems are likely to be the ultimate sink for a large fraction of nanomaterials. These terrestrial habitats may serve as sources to aquatic environments through runoff, erosion and intention release (Humphries *et al.*, 2006). This will likely create a new class of environmental pollutants that need to be assessed for their environmental fate and potential adverse effects to ecosystem and human health (Service, 2005).

Nanotechnology:

“It is the art and science of manipulating matter at the nanoscale (1 to 100 nm) to create new and unique materials and products with enormous potential to change society.” (National Nanotechnology Initiative)

Nanomaterials:

Materials with one or more components that have at least one dimension in the range of 1 to 100 nm and include nanoparticles, nanofibres and nanotubes, composite materials and nanostructured surfaces.



An original depiction showing two different approaches for the synthesis of nanoparticles:

(i) Bottom-up approach and (ii) Top-down approach

Generally, Nanoparticles are generated by two approaches which depend on the size and form of metal present in the solvent. For instance, if the size of precursors is less than the size of nano, *i.e.*, ions, atoms, or molecules, the pathway called “bottom-up” is operated. In this approach, the synthesis of Nanoparticles is achieved through the aggregation of molecules, atoms, and or different sized clusters through the reaction of chemical or biological reducing agents.

The other pathway is “top-down” which functions when the size of the precursor is larger than the size nano. Therefore, the particle size is substantially reduced to obtain the nano-size in this approach.

Potential sources of manufactured nanoparticles to the environment

Intentional routes

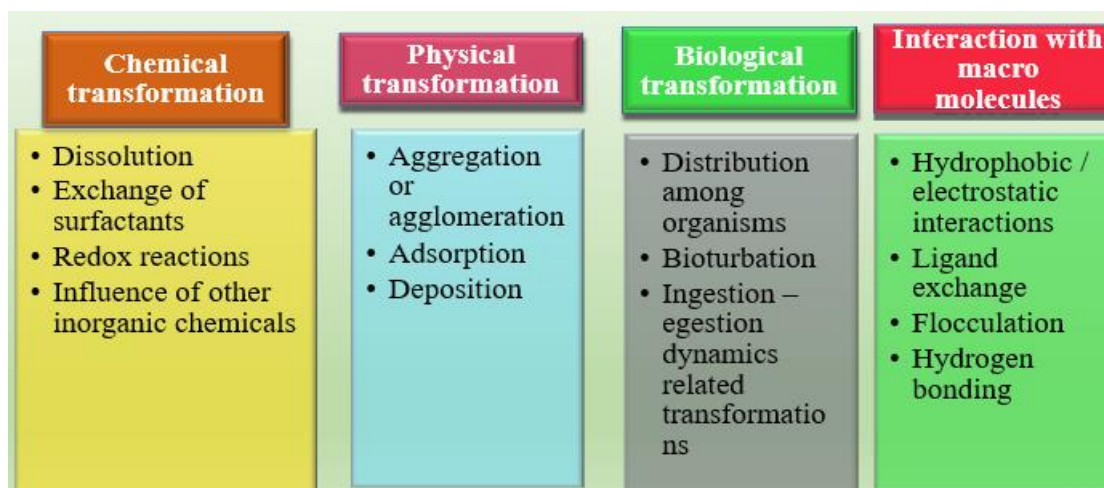
- Wastewater discharge – household, industry, waste water treatment plants

- Use of nano particles for environment cleanup (water cleanup), agriculture (nano fertilizer and pesticides)

Unintentional routes

- Accidental spills
- Consumption of products containing nanoparticles as additives
- Deposition from atmosphere due to air pollution

Environmental transformation of manufactured nano materials



Chemical Transformations

Dissolution. Dissolution is one of the transformation processes which can affect the fate, reactivity, toxicity, persistence and bioavailability of MeO (metal oxide)-NPs in the environment. Like other processes, dissolution of MeO-NPs also depends on their physicochemical properties and chemistry of the environmental system. Size and morphology of the MeO-NPs have a significant effect on their dissolution property due to the difference within surface area to mass ratio, smaller NPs have higher specific surface area than the larger ones

Redox reactions. Redox reaction comprises two coupled processes, oxidation and reduction reactions, which involve the transfer of electrons among the reacting species. MeO-NPs may compose of various constituents that undergo reduction and/or oxidation in environmental systems. The coating materials may also have significant effect on the redox reactions of the NPs.

Other chemical processes. The presence of other reactive organic and inorganic substances in the environment (e.g. surfactants capable of ligand exchange with NPs) can have a significant effect on the transformations and fate of MeO-NPs. If the system comprises reduced sulfur species, it can react with MeO-NPs and form various products in aquatic environments

Interaction with other inorganic chemicals: The presence of macromolecules such as NOM (natural organic matter), proteins, polysaccharides, and other surfactants in the environmental system may interact and transform the surface, and significantly alter the physicochemical properties of NPs. Thus, macromolecules can also determine fate of the NPs in the There are

various mechanisms proposed for interactions between the macromolecules and NPs (depend on the types and behavior of both materials) which include hydrophobic interaction, electrostatic and van der Waals interaction, chelation, ligand exchange, bridging, and hydrogen bonding, and these interaction processes are highly dependent on the pH of the medium

Physical Transformations

Aggregation/Agglomeration, adsorption, and deposition are the physical processes which have lasting implications on the fate, bioavailability, and toxicity of MeO-NPs

Deposition. Deposition, the behavior to settle on a bottom surface from a solution, is mainly favored by aggregation/agglomeration of the MeO-NPs. In general, aggregation, agglomeration and deposition processes are interrelated processes. Deposition increases when aggregation is higher, and eventual stability and deposition processes of MeONPs are likely determined by properties of the NPs (to some extent) and a combination of physicochemical parameters of the medium.

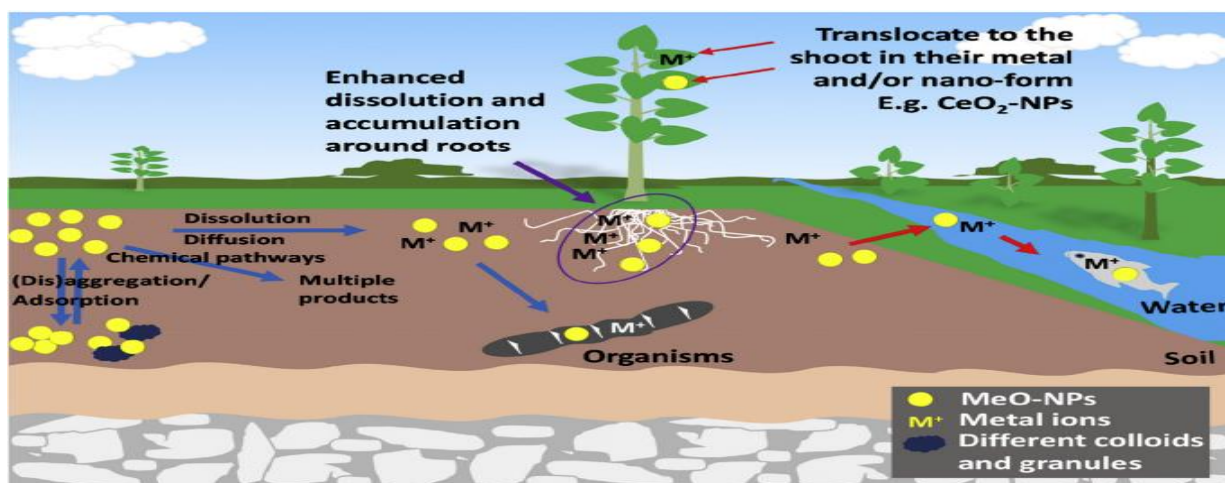
Nanotechnology in Agriculture

- ✓ Nano-agrochemical -Nano fertilizers
 - Nano herbicides
 - Nano fungicides
 - Nano insecticides
- ✓ Water management - Nano sensor

Agricultural applications of nanoparticles (NPs)

Nano materials	Applications
ZnO	Remediation and fortification of rice with low concentrations of Zn in soil
MnOx	Colorimetric nanosensor for indirect measurement of antioxidant capacity.
CuO, ZnO	Both NPs facilitate bifenthrin insecticide uptake in <i>Eisenia fetida</i> earthworms compared to only bifenthrin exposure.
MnO	Antifungal activity against soil-borne pathogens (<i>P. nicotianae</i> , <i>T. basicola</i>) with possibility to control other plant pathogens.
ZnO quantum dot	Sensor for pesticide detection in water
Zeolite/Fe ₂ O ₃	Nano fertilizer with less toxic effect toward humans compared to other fertilizers.
SiO ₂	Insecticide properties against leaf worm (<i>Spodoptera littoralis</i>).
Yb ₂ O ₃	Fluorescent sensor for imazapyr herbicide detection

Bioavailability of Nano Materials



Common transformations, transport and bioavailability of manufactured NPs in a terrestrial environment

The common mechanisms such as passive diffusion, facilitated diffusion, active transport and endocytosis proposed for the cellular internalization of any materials may also be the pathways for the uptake of MeO-NPs and their transformation products, and the importance of these ways are strongly dependent on the type of the organisms. Bioavailability of MeO-NPs depends on their stability in the environment and stable NPs are less bioavailable and also exhibit lower toxic-effect to cellular organisms as their interactions with plasma proteins are important upon introduction of the materials. After the uptake, NPs and its other forms may undergo various transformations like aggregation, dissolution, complexation, reduction, and others

Bioavailability of nanomaterials refers to the extent and rate at which they can be absorbed, distributed, metabolized, and excreted by organisms or systems within the environment. In the context of agroecosystems, bioavailability plays a crucial role in determining the uptake of nanomaterials by plants, soil organisms, and other components of the ecosystem.

Several factors influence the bioavailability of nanomaterials in agroecosystems:

Particle Size and Surface Chemistry: Nanomaterials exhibit unique physicochemical properties that can influence their interaction with biological systems. Smaller nanoparticles generally have higher bioavailability due to their larger surface area-to-volume ratio, which enhances interactions with biological molecules and surfaces. Surface chemistry, including surface charge, functionalization, and coatings, also affects the behavior and bioavailability of nanomaterials.

Environmental Conditions: Environmental factors such as pH, temperature, humidity, and the presence of organic matter can significantly influence the fate and bioavailability of nanomaterials in agroecosystems. Changes in environmental conditions may alter the

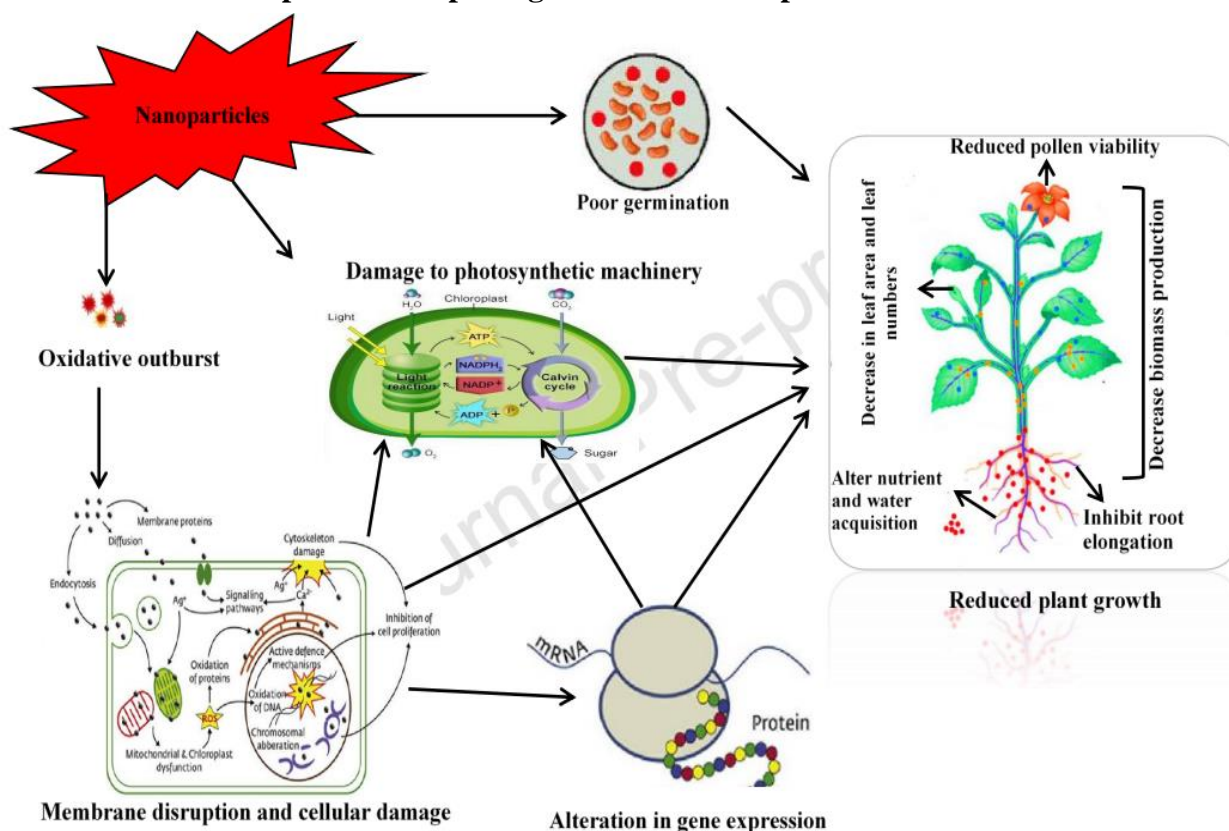
aggregation, dissolution, or transformation of nanomaterials, thereby affecting their bioavailability to organisms.

Soil Properties: Soil characteristics, such as texture, organic matter content, and microbial activity, can impact the mobility and bioavailability of nanomaterials in soil environments. Nanoparticles may interact with soil components, adsorb onto soil surfaces, or be taken up by soil organisms, affecting their bioavailability to plants and other organisms in the soil food web.

Plant Uptake Mechanisms: Plants can uptake nanomaterials through various pathways, including root uptake, foliar absorption, and uptake through symbiotic associations with mycorrhizal fungi. The physicochemical properties of nanomaterials, such as size, surface charge, and surface functionalization, influence their uptake and translocation within plants. Once inside the plant, nanomaterials may accumulate in different plant tissues, affecting plant growth, development, and nutritional quality.

Understanding the bioavailability of nanomaterials in agroecosystems is essential for assessing their potential risks and benefits in agriculture. It enables researchers to evaluate the environmental fate, ecological impacts, and human health implications of nanomaterials, thereby guiding the development of sustainable nanotechnology applications in agriculture.

Toxic effects of nanoparticles on plant growth and development



The higher concentration of nanoparticles causes alteration in morphology and physiological processes of crop plants. Higher concentration of nanoparticles in root zone inhibits seed germination, restricts root development; alters water and nutrient uptake, and decreases leaf development and biomass production. Moreover, nanomaterial toxicity causes

oxidative outburst resulting in chloroplast disorganization, reduced photosynthesis, membrane disruption, cellular damage and altered gene expression. Toxic effects of nanoparticles on plant growth and development. The higher concentration of nanoparticles causes alteration in morphology and physiological processes of crop plants. Higher concentration of nanoparticles in root zone inhibits seed germination, restricts root development; alters water and nutrient uptake, and decreases leaf development and biomass production. Moreover, nanomaterial toxicity causes oxidative outburst resulting in chloroplast disorganization, reduced photosynthesis, membrane disruption, cellular damage and altered gene expression.

Phytotoxic effect of nano particles in crops

Nanoparticles	Crop	Phytotoxic effects observed	References
Zinc and zinc oxide	Radish, rape grass and rye grass	Inhibition of root growth and decrease in shoot biomass	Ocsoy <i>et al.</i> , (2013)
	Onion	Affect cellular metabolism – cell division	Kumar <i>et al.</i> , (2010)
Silver	Onion	Cytotoxic and genotoxic impacts in root tip	Lamsal <i>et al.</i> , (2011)
	Rice	Severe damage in metabolism, damaged cell wall and vacuoles	Jo <i>et al.</i> , (2009)
Carbon nano tubes	Tomato	Accumulation of ROS causes cell death, change in gene expression in roots, leaves and fruits	Qian <i>et al.</i> , (2011)
Titanium dioxide	Corn, Soyabean, cabbage and carrot	Inhibited the root elongation	Pinto <i>et al.</i> , (2013)
Alumina	Cowpea	Swelling of roots	Seo <i>et al.</i> , (2011)

The toxicity of nanomaterials in crop ecosystems is a significant concern due to their potential adverse effects on plant health, soil quality, and ecosystem functioning. Nanomaterials can exert toxicity through various mechanisms, including direct cytotoxicity, oxidative stress, disruption of cellular processes, and alteration of soil microbial communities. Understanding the toxicity of nanomaterials in crop ecosystems is crucial for assessing their environmental risks and developing strategies to minimize their impact.

Several factors contribute to the toxicity of nanomaterials in crop ecosystems:

Nanoparticle Characteristics: The physicochemical properties of nanoparticles, such as size, shape, surface charge, and composition, influence their toxicity to crops. Smaller nanoparticles generally have higher toxicity due to their increased surface area and reactivity. Surface modifications, such as functionalization or coating, can also affect the toxicity of nanoparticles by altering their interaction with plant cells and soil components.

1. **Nanoparticle Characteristics:** The physicochemical properties of nanoparticles, such as size, shape, surface charge, and composition, influence their toxicity to crops. Smaller nanoparticles generally have higher toxicity due to their increased surface area and reactivity. Surface modifications, such as functionalization or coating, can also affect the toxicity of nanoparticles by altering their interaction with plant cells and soil components.
2. **Uptake and Translocation:** Nanoparticles can enter plants through root uptake, foliar absorption, or association with soil microorganisms. Once inside the plant, nanoparticles may translocate to different tissues, including roots, stems, leaves, and fruits. The uptake and translocation of nanoparticles can disrupt cellular processes, interfere with nutrient uptake, and impair plant growth and development.
3. **Oxidative Stress:** Nanoparticles can induce oxidative stress in plants by generating reactive oxygen species (ROS), such as superoxide radicals, hydrogen peroxide, and hydroxyl radicals. Excessive ROS production can damage cellular components, including lipids, proteins, and DNA, leading to cell death and tissue damage. Plants respond to oxidative stress by activating antioxidant defense mechanisms, but prolonged exposure to nanoparticles can overwhelm these defenses, resulting in oxidative damage.
4. **Soil Health:** Nanoparticles released into the soil can affect soil microbial communities and soil biogeochemical processes. Nanoparticles may inhibit soil microbial activity, disrupt nutrient cycling, and alter soil structure and fertility. Changes in soil health can impact crop growth and productivity, as well as the overall ecosystem functioning.

Toxicity assessments of nanomaterials in crop ecosystems typically involve laboratory studies, greenhouse experiments, and field trials to evaluate their effects on plant growth, morphology, physiology, and yield. Researchers employ a range of techniques, including molecular biology, microscopy, spectroscopy, and bioinformatics, to elucidate the mechanisms underlying nanomaterial toxicity and its implications for crop production and environmental sustainability.

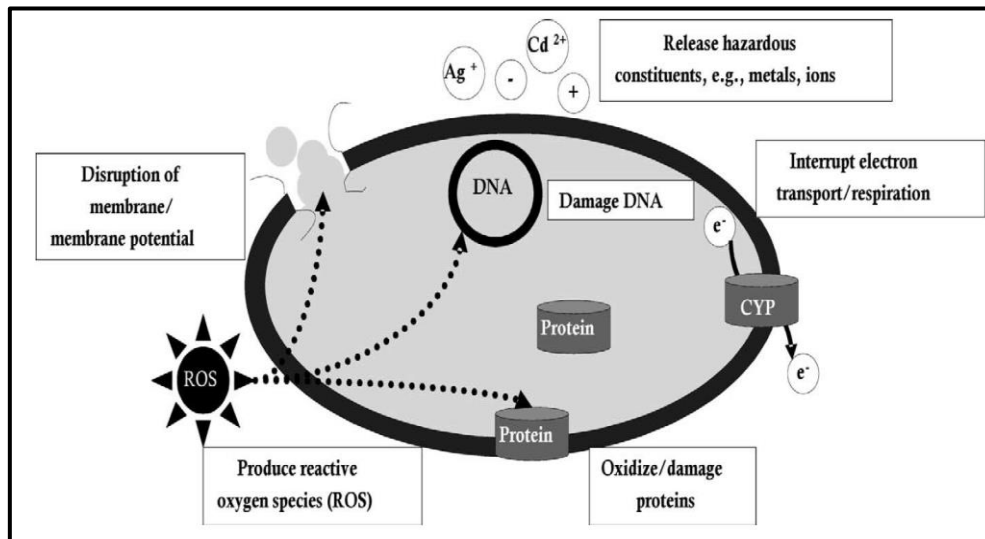
Overall, addressing the toxicity of nanomaterials in crop ecosystems requires a multidisciplinary approach that integrates nanoscience, plant biology, soil science, and environmental toxicology. By understanding the mechanisms of nanomaterial toxicity and its ecological consequences, scientists can develop safe and sustainable nanotechnology solutions for agriculture.

Toxicity of nanomaterials to soil microbes

Application of nanomaterials to soil have the impact on soil microorganisms due to some of the mechanisms like cell membrane disruption, production of reactive oxygen species, damage to the proteins or through interruption of electron transport and respiration

The toxicity of nanomaterials to soil microbes is a critical aspect of their environmental impact, as soil microorganisms play key roles in nutrient cycling, soil fertility, and ecosystem functioning. Nanomaterials can interact with soil microbes through direct contact, release of ions or reactive oxygen species (ROS), and alteration of soil physicochemical properties.

Understanding the toxicity of nanomaterials to soil microbes is essential for assessing their environmental risks and developing strategies to mitigate their impact on soil health and ecosystem services.



Possible mechanisms of nanomaterial toxicity to bacteria. Different nanomaterials may cause toxicity via one or more of these mechanisms

Several factors contribute to the toxicity of nanomaterials to soil microbes:

Several factors contribute to the toxicity of nanomaterials to soil 5

1. **Nanoparticle Characteristics:** The physicochemical properties of nanoparticles, such as size, shape, surface charge, and composition, influence their toxicity to soil microbes. Smaller nanoparticles generally have higher toxicity due to their larger surface area-to-volume ratio and increased reactivity. Surface modifications, such as functionalization or coating, can also affect the interaction of nanoparticles with soil microbes.
2. **Disruption of Cellular Processes:** Nanoparticles can disrupt essential cellular processes in soil microbes, including cell membrane integrity, enzyme activity, and metabolic pathways. Nanoparticles may penetrate microbial cell walls, leading to membrane damage and leakage of cellular contents. Additionally, nanoparticles can interfere with enzymatic reactions involved in nutrient cycling and energy metabolism, impairing microbial growth and activity.
3. **Generation of Reactive Oxygen Species (ROS):** Some nanoparticles have the ability to generate ROS, such as superoxide radicals, hydrogen peroxide, and hydroxyl radicals, through processes such as photoactivation or chemical reactions with environmental constituents. Excessive ROS production can cause oxidative stress in soil microbes, leading to damage to biomolecules such as lipids, proteins, and DNA.
4. **Release of Metal Ions:** Nanoparticles composed of metals or metal oxides can release metal ions into the soil environment through dissolution or corrosion processes. Metal ions

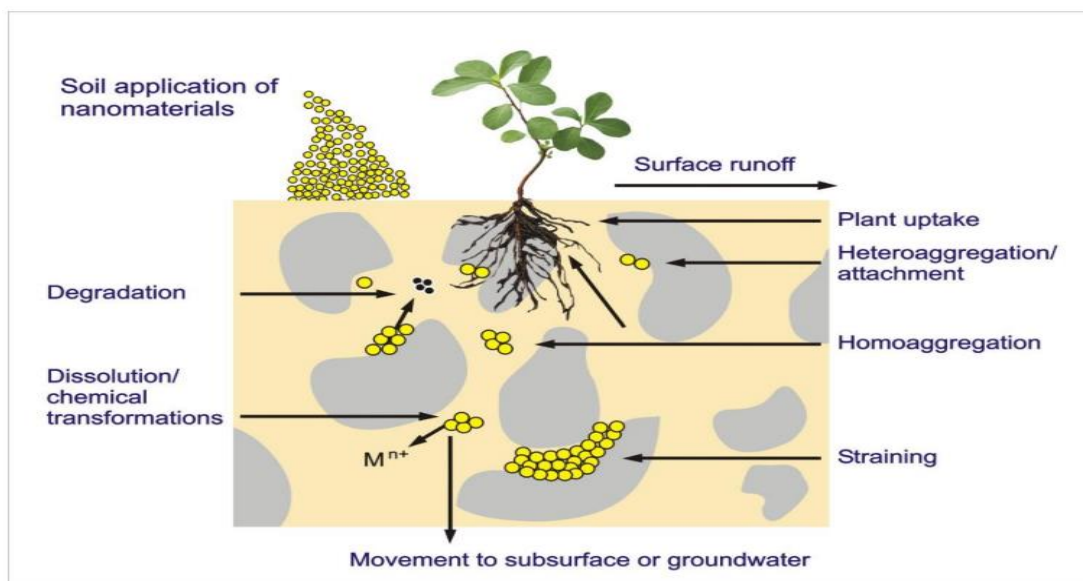
released from nanoparticles may exhibit toxic effects on soil microbes by interfering with cellular processes, disrupting enzyme activities, and inducing oxidative stress.

5. **Alteration of Soil Properties:** Nanomaterials can alter soil physicochemical properties, such as pH, organic matter content, and soil structure, which in turn can affect microbial communities and microbial activity. Changes in soil properties may impact microbial diversity, abundance, and function, ultimately influencing nutrient cycling, soil fertility, and plant-microbe interactions.

Toxicity assessments of nanomaterials to soil microbes typically involve laboratory experiments using model microorganisms or soil microbial communities. Researchers employ a variety of techniques, including microbiological assays, molecular biology methods, and omics technologies, to evaluate the effects of nanomaterial exposure on microbial growth, viability, metabolic activity, and community composition.

Overall, addressing the toxicity of nanomaterials to soil microbes requires a comprehensive understanding of the interactions between nanomaterials and soil microbial communities, as well as their implications for soil health and ecosystem functioning. By elucidating the mechanisms of nanomaterial toxicity to soil microbes, scientists can develop strategies to minimize the environmental risks associated with nanotechnology applications in agriculture and environmental remediation.

Fate of Nanomaterials



The fate of nanomaterials in soil systems is a complex and multifaceted topic that has garnered significant attention in recent years due to the increasing use of nanomaterials in various industries and their potential environmental impacts. Understanding how nanomaterials behave in soil environments is crucial for assessing their potential risks to ecosystems and human health. Here are some key points regarding the fate of nanomaterials in soil systems:

Transport: Nanomaterials can be transported through soil via various mechanisms such as advection, diffusion, and plant uptake. The mobility of nanomaterials in soil depends on factors such as particle size, surface chemistry, soil properties, and environmental conditions.

Retention: Nanomaterials may adhere to soil particles through physical and chemical interactions,

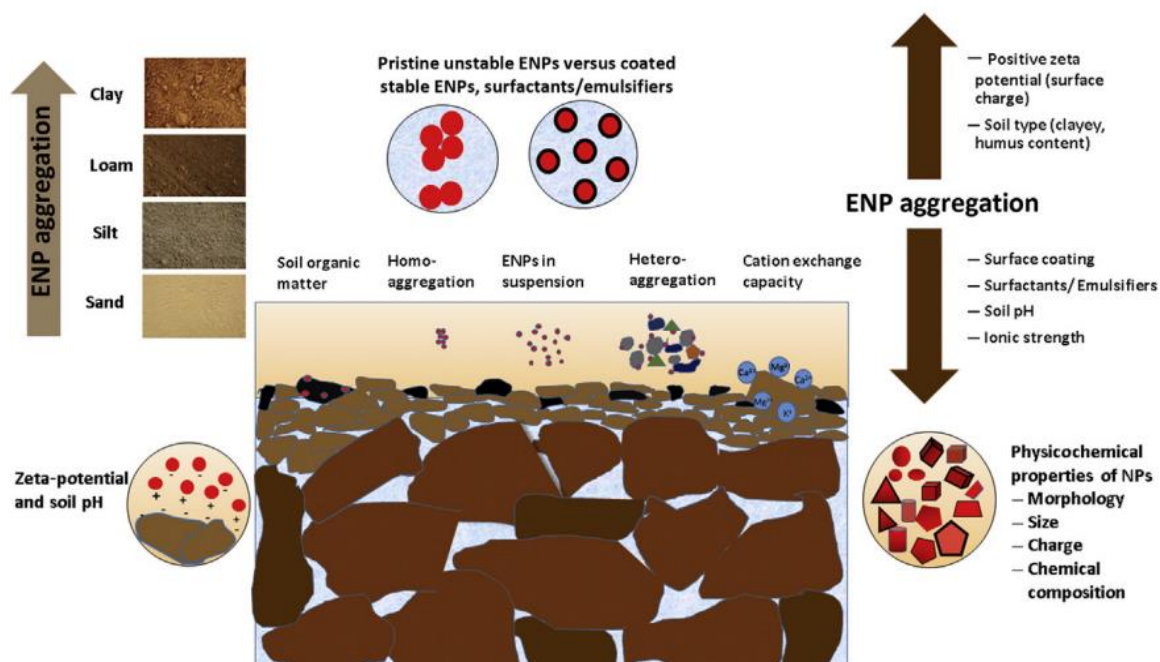
leading to their retention in the soil matrix. The extent of retention depends on factors such as surface charge, surface area, and soil properties (e.g., organic matter content, clay content).

Transformation: Nanomaterials in soil may undergo physical, chemical, and biological transformations over time. Processes such as aggregation, dissolution, surface oxidation, and microbial degradation can alter the properties and behavior of nanomaterials in soil environments.

Bioavailability: Nanomaterials can interact with soil organisms and plants, potentially affecting their bioavailability and toxicity. Some nanomaterials may be taken up by plants and subsequently enter the food chain, posing risks to organisms at higher trophic levels.

Overall, the fate of nanomaterials in soil systems is influenced by a complex interplay of physical, chemical, and biological processes. Further research is essential to improve our understanding of the environmental behavior and potential risks associated with nanomaterials in soils. Regulatory frameworks may need to be developed or adapted to address the potential environmental implications of nanomaterial use in various industries.

Factors influencing the fate, transport, and retention of ENMs in soil.



Nanomaterial Properties: The physical and chemical characteristics of nanomaterials play a significant role in their behavior in soil. Properties such as particle size, shape, surface charge, surface chemistry, and surface coating can affect interactions with soil particles and organisms.

Soil Properties: Soil characteristics such as texture, organic matter content, pH, clay mineralogy, and moisture content influence the fate and transport of nanomaterials. For example, soils with high clay content tend to have greater adsorption capacity for nanomaterials, while organic matter can affect their stability and mobility.

Environmental Conditions: Environmental factors such as temperature, humidity, rainfall, and redox potential can affect the behavior of nanomaterials in soil. Changes in environmental conditions may influence processes such as aggregation, dissolution, and microbial activity, thereby impacting the fate of nanomaterials in soil systems.

Soil Structure: Soil structure, including pore size distribution, porosity, and soil aggregation, affects the movement of nanomaterials within the soil matrix. Pores of different sizes may facilitate or hinder the transport of nanomaterials, while soil aggregation can affect their retention and release.

Biological Factors: Soil microorganisms, plants, and soil fauna can interact with nanomaterials and influence their fate in soil. Microbial activity may degrade or transform nanomaterials, while plant roots can facilitate their uptake and translocation. Soil organisms such as earthworms and nematodes can also affect the distribution and fate of nanomaterials in soil.

Understanding how these factors interact to influence the fate, transport, and retention of nanomaterials in soil is essential for assessing their environmental risks and developing strategies to mitigate potential impacts. Further research is needed to improve our understanding of these complex interactions and their implications for soil health and ecosystem integrity.

In conclusion, the bioavailability, toxicity, and fate of nanomaterials in crop ecosystems are multifaceted and interconnected phenomena that necessitate careful consideration for sustainable agricultural practices. Nanotechnology holds immense potential for enhancing crop productivity, nutrient delivery, and pest management, but its widespread adoption requires a thorough understanding of its environmental implications.

Conclusion:

Throughout this book chapter, we have explored the complex interactions between nanomaterials and crop ecosystems, highlighting key findings and challenges:

1. **Bioavailability:** The bioavailability of nanomaterials is influenced by their physicochemical properties, environmental conditions, and interactions with soil and plant components. Understanding the mechanisms governing the uptake and translocation of nanomaterials in crops is essential for predicting their environmental fate and assessing their potential risks to human health and ecosystems.
2. **Toxicity:** Nanomaterial toxicity in crop ecosystems can arise from direct cytotoxic effects, oxidative stress, disruption of cellular processes, and alteration of soil microbial communities. Assessing the toxicity of nanomaterials to crops, soil microbes, and other non-target organisms is critical for evaluating their safety and minimizing unintended environmental consequences.

3. **Fate:** The fate of nanomaterials in crop ecosystems involves their degradation, accumulation, and persistence in soil, water, and plant tissues. Nanomaterials may undergo transformation processes, such as dissolution, aggregation, or surface modifications, which can influence their environmental behavior and long-term impacts on soil health and ecosystem functioning.

In light of these findings, several considerations and future directions emerge:

- **Risk Assessment:** Robust risk assessment frameworks are needed to evaluate the environmental risks associated with nanomaterials in crop ecosystems. This requires standardized toxicity testing protocols, predictive modeling approaches, and multi-stakeholder engagement to ensure the safe and responsible deployment of nanotechnology in agriculture.
- **Sustainable Nanotechnology:** Developing sustainable nanotechnology solutions involves minimizing the environmental footprint of nanomaterials through green synthesis methods, eco-friendly disposal strategies, and the design of biodegradable nanomaterials with reduced toxicity and environmental persistence.
- **Knowledge Gaps:** Addressing knowledge gaps in nanomaterial behavior, fate, and toxicity in crop ecosystems requires interdisciplinary research efforts spanning nanoscience, plant biology, soil science, environmental toxicology, and agricultural engineering. Collaborative research initiatives and data-sharing platforms can facilitate knowledge exchange and inform evidence-based decision-making.

Overall, by integrating scientific knowledge, technological innovation, and stakeholder engagement, we can harness the potential of nanotechnology to advance sustainable agriculture while safeguarding the health of ecosystems and communities. Embracing a precautionary approach and adopting principles of responsible innovation will be crucial in realizing the promise of nanotechnology for a resilient and thriving agricultural future.

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SYNTHETIC SEED PRODUCTION

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Abstract:

The seed is a functional element of sexual reproduction of higher plant. In nature, the humble beginning of the independent life of higher plants starts along with seed germination. Seeds are the “mysterious genetic capsules” which store the genetic information and carry forward to next progeny. The zygotic embryo present inside the botanical seed serves as propagules to produce offspring, and these embryos are always heterozygous because of the recombination during meiotic crossing over in the course of gamete formation as well as for mix-up of the genome of two different parents through cross-pollination. In seed-propagated crops, the agricultural yield is highly unstable due to heterozygosity among seed-derived plants. The answer of this problem is synthetic seeds- the functional mimic of botanical seeds.

Introduction:

In nature, seeds are typically the primary method of plant propagation. Several crops have failed at seed propagation due to factors like heterozygosity, small size, lack of endosperms, and requirement for fungal infection for germination. (Saiprasad, 2001). Artificial seeds are encapsulation of somatic embryos, shoot bud, aggregates of cell of any tissues in gel matrix under aseptic condition in a sterile environment later on which can be sown into a plant either in in-vitro or in-vivo condition. The somatic embryo can be encapsulated, handled and used like a natural seed was first suggested by Murashige (1977) and efforts to engineer them into synthetic seed have been ongoing ever since (Gray, 1987). Bapat *et al.*, (1987) proposed the encapsulation of shoot tip in *Morus indica*; these applications have made the concept of synthetic seed set free from its bonds to somatic embryos and broaden the technology to the encapsulation of various *in vitro* derived propagules. He defined artificial seeds as “an encapsulated single somatic embryo”. Later on, Gray *et al.* defined it as “a somatic embryo that is engineered for the practical use in commercial plant production”. As artificial seed have ability to extend its period of storage, they can eliminate the acclimation steps necessary in micro propagation and give breeders greater flexibility. The term, “EMBLING” is used for the plants originated from synthetic seed.

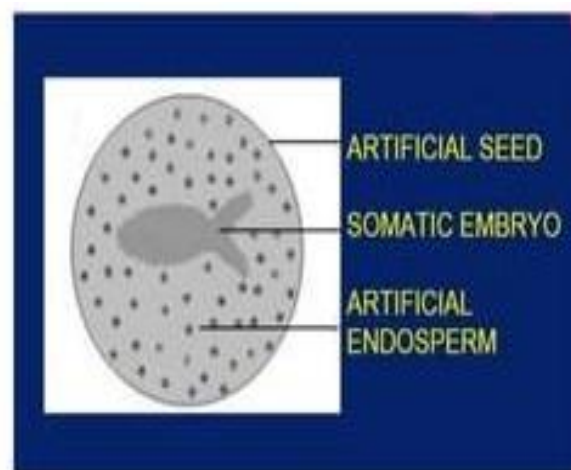
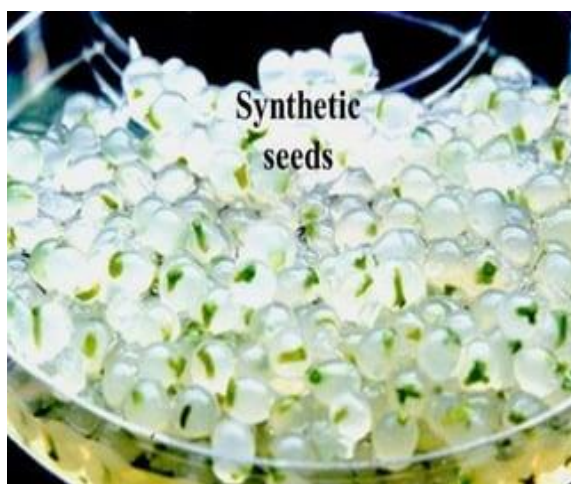
Why Synthetic Seeds?

One of the reasons is that this technology can help in improving characteristics in the new embryos and hence can be beneficial for longer duration storage. For instance, these embryos and plant propagules are prone to weed infestations as well as pathogen infections that can affect their ability to survive. Therefore, adding herbicides or biological pesticides along with nutritive

material provides resistance against weeds and even pests for the initial growth phase, thus, facilitating better growth and development. Another important reason is to prevent the desiccation of somatic embryos or plant propagules in the natural environment. Hence, to prevent these outcomes, protective coatings are suggested for somatic embryos or propagules. They are also important for some seedless plant species or varieties, for instance, grapes where it is hard to have successful propagation via seeds.

Synthetic Seed Structure

The structure of the synthetic seed is similar to that of the natural seed. It is made up of explant material, which resembles the zygotic embryo in a conventional seed, and the capsule, which consists of a gel agent and additional ingredients like nutrient content, growth hormones, anti-pathogens, bio-controllers, and bio-fertilizers and imitates the endosperm in a conventional seed (Fig. 1).



Types of Artificial (Synthetic) Seeds

1. Desiccated Artificial Seeds

These types of seeds are achieved from somatic embryos either naked or encapsulated in polyoxyethylene glycol followed by their desiccation. Desiccation can be applied either rapidly by leaving artificial seeds in unsealed petri dishes on the bench overnight to dry, or slowly over a more controlled period of reducing relative humidity (Ara *et al.*, 2000). These types of artificial seeds can be only made in plants whose somatic embryos are desiccation-tolerant (Sharma *et al.* 2013). This type of synthetic seeds is produced in desiccation tolerant plant species such as *craterstigma lantagineum*, *linderni abbrevidens*, *ramonda serbica*.

2. Hydrated Synthetic Seeds:

Hydrated synthetic seeds are produced by encapsulating the somatic embryos in hydrogel-like sodium alginate, potassium alginate, carrageenan, sodium pectate or sodium alginate with gelatine. It is the most studied and best method to supply protection and to convert their *in vitro* micropropagules into 'artificial seeds' or 'synseeds' artificial seed production (Redenbaugh, 1993). Such seeds are produced in those plants where somatic embryos are

recalcitrant and sensitive to desiccation (Ara *et al.*, 2000). Calcium alginate gel is most suitable and used gel.

Propagules for the Formation of Synseeds-

Earlier, only somatic embryos were used as explants for the formation of synthetic seeds in a number of plants. However, later reports by various researchers revealed the use of a variety of plant micro propagules, including monopolar axillary shoot tips as well as buds, nodal segments, embryogenic masses, and calli, along with a variety of other explants, including bulb, bulblets, hairy roots, microtubes, and protocorms or protocorm-like bodies (Reddy *et al.*, 2012; Gantait & Sinniah, 2012).

I. Somatic Embryo

Somatic embryos are those that are created asexually through somatic cells alone without union of gametes. Somatic embryos are the most common micro propagule used for artificial seed production because their structures are able to produce the radical and plumule axis, which has the capability to progress into the root and shoot in a single step (Ara *et al.*, 2000). However, unlike normal embryos, SEs do not experience desiccation or dormancy and instead begin to germinate as soon as they are completely developed (Zimmerman, 1993).

II. Calli and Embryogenic Masses

For the development of clonal plants and for the investigation of genetic change, regenerative and stable embryogenic masses can be employed. However, due to repeated subculturing, keeping them for a longer period of time in bioreactors and culture containers is challenging (Ara *et al.*, 2000). Encapsulating these embryogenic masses in sodium alginate and storing them at 40°C following 6-benzyl amino purine (BAP) treatment might avoid the time-consuming and costly subculturing method (Redenbaugh *et al.*, 1991). Calli's undifferentiated character and minimal differentiation capability restrict their adoption for explant propagules for the manufacture of synseed (Gantait *et al.*, 2015). For the first time, the use of calli for the formation of synseeds was successfully observed in *Allium sativum* (Kim & Park, 2002), exhibiting a high rate of conversion and regeneration of synseeds to plants.

III. Apical Shoot Tips/ Shoot Buds and Nodal Segment

In order to create synthetic seeds, auxiliary shoot buds and/or apical shoot tips that lack root meristems have also been encapsulated. When compared to micro propagation of shoot tip/buds encapsulated synseeds, traditional shoot tip culture *in vitro* takes more space and culture medium (Gantait *et al.*, 2015). The need for less space means that moving plant propagules from one location to another is simple.

Steps in Production of Synthetic Seed

The Production of Commercial Synthetic Seeds Requires the Following Steps

1. Creation of somatic embryos tissue from tissue or cell transformation.
2. Synchronous somatic embryo formation on a large scale.
3. The somatic embryo's maturation.
4. Process for non-toxic encapsulation or coating.
5. Depending on the species, artificial endosperm or a giant gametophyte.

6. The capacity of synthetic seeds to store food.
7. Depending on output, high frequency direct plant conversion from greenhouse or nursery field requirements

Encapsulation Methods for Synthetic Seeds

A. Dropping Method

Prepare 1-5% sodium alginate solution and insert somatic embryos in the prepared gel. Then take this sodium alginate gel containing somatic embryos in the funnel and drip the drops of gel along with somatic embryos from the tip of funnel to the prepared known concentration of complex solution containing calcium chloride or calcium salts used as complexing agents. Keep the gel containing somatic embryos in complex solution for 20 minutes. After forming the gel capsules wash it in water for 5 minutes (Saiprasad, 2001; Lambardi *et al.*, 2006).

B. Molding Method:

In molding method, a simple procedure is followed. In this, temperature dependent gels are used (such as agar) and embryos are mixed in these gels. At low temperature, coating of cells with gel takes place.

Procedure for Artificial Seed Production

Selection of Plant Material: Select the plant material which is disease free, fresh, and can multiply or induce very fast and can give embryogenic induction.

Slice Explant: Excise roots, stem, leaves, bud, embryo sac, nucellus *etc* as explants.

Sterilization of Explant: Different chemicals used for surface sterilization of explants are as given in the table below:

Table 1: List of Chemicals with concentration used for surface sterilization of explants

Disinfectant	Product No.	Concentration (%)	Exposure (min)
Calcium hypochlorite	211389	9-10	5-30
Sodium hypochlorite*	425044	0.5-5	5-30
Hydrogen peroxide	H1009	3-12	5-15
Ethyl alcohol	E7148	70-95	0.1-5.0
Silver nitrate	S7276	1	5-30
Mercuric chloride	M1136	0.1-1.0	2-10
Benzalkonium Chloride	B1383	0.01-0.1	5-20

Stock Solution Preparation: Prepare stock of macro nutrient, micronutrient, vitamins, growth hormones.

Preparation of MS media: Add all the stocks like macro, micro, vitamin, hormones, agar as solidifying agent and sucrose as a source of carbon. Maintain pH and autoclave.

Inoculation of Explant: Inoculate the sterilized explants into petri jars inside laminar air flow.

Induction of Callus on Nutrient Medium: Induction of callus in nutrient medium in growth room.

Maintenance of Medium: Maintenance of medium is done to avoid damage of cells or tissue.

Transfer of Callus in Suspension Culture: Depending upon plant species callus should transfer either to another embryogenic induced medium like MS or should transfer to suspension culture with given hormonal combination for that species for inducing embryo developmental stages.

Embryo Differentiation: Differentiation of embryo into different stages.

Excision of Embryo: Embryo is excised from nutrient culture medium

Preparation of Gel Matrix: Prepare (1-5%) sodium alginate gel or any required gel in known concentration.

Insertion of Somatic Embryos: Insert excised somatic embryos in sodium alginate solution and mixed it.

Preparation of Complex Solution: Complex solution is prepared at the con of 30-100uM, where calcium salts or calcium chloride mostly used as complexing agents.

Dropping Method of Encapsulation: Take sodium alginate gel containing somatic embryos in the funnel and drip the drops of gel along with somatic embryos from the tip of funnel to the prepared known concentration of complex solution containing calcium chloride or calcium salts used as complexing agents. Keeps the gel containing somatic embryos in complex solution for 20 minutes. After forming the gel capsules wash it in water for 5 minutes.

Table 2: Encapsulation materials for synthetic seed

Gel concentration (% w/v)	Complexing agents	Concentration ion (uM)
Sodium alginate (0.5-5.0)	Calcium salts	30-100
Sodium alginate (2.0) with Gelatin (5.0)	Calcium chloride	30-100
Carrageenan (0.2-0.8)	Potassium chloride Ammonium chloride	500
Locust bean gum (0.4-1.0)	Potassium chloride Ammonium chloride	500
Gelrite (0.25)		

Addition of Adjuvants to the Matrix

Macro nutrients, micronutrients, growth regulators, vitamins, sucrose as a source of carbon or a number of useful materials such as fungicides, pesticides, antibiotics and microorganisms (eg. rhizobia) may be incorporated into the encapsulation matrix to prevent embryo from desiccation state. Incorporation of activated charcoal into the gel matrix also improves the conversion and vigour of the encapsulated somatic embryos and it also retain the nutrients within the hydrogel capsule that results in slowly releases them to the growing embryo inside gel.

Cryopreservation of Synthetic Seeds

Cryopreservation is a technique for long-term storage of plant germplasm for next generations. Cryopreservation can be done at ultra-low temperature at -196 C in liquid nitrogen. The major advantage of this technique is require minimum space, rare of contamination, no need of in-vitro culture can directly grow outside the environment, stored for long period of time. The long-term conservation of embryogenic cell lines may be a valuable tool in genetic engineering.

Encapsulations of seed in calcium alginate gel in in vitro or in in vivo generated explants are proved to be reliable with propagation and cryo storage (Verleysen *et al.*, 2005).

Advantages

Merits of artificial or synthetic seed are listed below.

1. Artificial seeds are reasonably inexpensive to produce and easy to handle, plant, and transport. They can also be stored for a long period using dehydration and cryopreservation techniques (Pennycooke and Towill, 2001; Wang, 2002).
2. An important technique for transgenic plants, where a single gene can be placed in a somatic cell and then this gene will be located in all the plants produced from this cell. Therefore, artificial seeds could be an efficient technology used for reproduction of transgenic (Daud, 2008).
3. To avoid extinction of endangered species and seedless plants: The most important advantages of synthetic seed are such as it helps in conservation of endangered species e. g. in hedgehog cacti (*Echinocereus* sp.) and seedless varieties e.g. grapes.
4. Large scale propagation: After the standardization of protocol it is very much suitable for large scale monoculture.
5. Germplasm conservation: A synthetic seed plays an important role in germplasm conservation.
6. Elite plant genotypes: artificial seed technology preserves, protects and permits economical mass propagation of elite plant genotypes such as orchids.
7. Independent of environmental conditions: The technologies of synthetic seeds production are not a season dependent as these are prepared inside the laboratory.
8. Permits direct field use: For tissue culture raised plants rooting, hardening is necessary but in case of synthetic seeds direct field sowing can give good yield.
9. Supply of beneficial adjuvants: beneficial adjuvants like plant nutrients, plant growth regulators, microorganisms, fungicides, mycorrhizae, antibiotics can be made available to the developing plant embryo as per the requirement as these can be added in to the matrix.
10. Hybrid production: synthetic seed production technology can be used for production of hybrids which have unstable genotypes or show seed sterility such as not susceptible towards infection. It can be used in combination with embryo rescue technique. The rescued embryo can be encapsulated with this technique to form synthetic seeds (Saiprasad, 2001).
11. Artificial seed production is an essential technique for the proliferation of plant species which are not able to produce seed, such as seedless grapes and seedless watermelon (Saiprasad, 2001).

Disadvantages

Several intensive researches in the field of synthetic seed technology were applied in propagating and conserving a number of plant species, but practical implementation of the technology is limited due to the following main reasons.

1. The technology of synthetic seed is limited due to poor conversion of even apparently normally matured somatic embryos and other micro propagules into plantlets (Ara *et al.*, 2000).
2. Limitations in storage caused by lack of dormancy, synchronic deficiency in somatic embryo development, improper maturation, low levels of conversion into plantlets, limitation in production of viable mature somatic embryos (Reddy *et al.*, 2012).
3. The reduction of viability and plant recovery when the artificial seeds are stored at low temperature (Makowczy and Andrzejewska, 2006).
4. The difficulties of sowing artificial seeds directly in soil or in commercial substrates such as compost, vermiculite, etc., under non-sterile conditions are considered to be one of the main limitations of the practical use of this technique (Rihan *et al.*, 2012).

Conclusion:

The most important requirement about the practical application of artificial seed is the production of high quality micropropagules and large-scale production of seeds. Among tree species, regeneration of viable plantlets from somatic embryos is a frequently encountered problem, including germination, maturation, rooting of shoots or acclimatization and shoot apex elongation. Occurrence of high levels of somaclonal variations in tissue culture is another aspect to be considered seriously while recommending the use of artificial seeds for clonal propagation. Commercialization of synthetic seeds still requires some additional processes such as improvement in the tissue culture practices for the generation of adequate propagules. The production of synthetic seed must either reduce production cost or increase crop value. However further study and research related to the improvement of technology can help in the global acceptance of synthetic seeds.

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TRANSFORMING RURAL LIVELIHOODS IN INDIA THROUGH THE USE OF AI IN AGRICULTURE

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Introduction:

Agriculture lies at the core of the Indian economy, continuing to feed its teeming population and being the source of livelihood for many. It contributes around 15% to the nation's GDP. Prior to the Green Revolution that began in the 1960s, India was heavily dependent on the import of food grains to meet the food requirement of its population. The situation was aggravated due to the Great Bengal Famine of the 1940s which stagnated the rice yields, affecting the agriculture dependent population. Although Green Revolution enabled the nation to overcome agricultural distress to a large extent, its complete eradication is rather complex. A multiple number of factors like fluctuating agricultural growth rates, unpredictable weather patterns, particularly monsoon rainfall behavior, inefficient agricultural markets, etc. are some of the main causes behind income variability among farmers (Anon., 2018). In the previous decades, agricultural sector has undergone a variety of technological revolutions to bring significant improvements in crop and animal production. The first agricultural revolution represented a transition from hunting and gathering to settled agriculture, while the second and third agricultural revolutions were associated with application of improved technology through mechanization (Rose and Chilvers, 2018), thereby taking the agricultural exports to a record high of USD 50 billion in 2021-22 (Anon., 2022). Currently, smart technologies, such as Artificial Intelligence (AI), robotics, cloud computing and the Internet of Things (IoT), has made its way into the agriculture sector, better known as Agriculture 4.0, aimed at enhancing productivity and achieving greater eco-efficiency. Advent of AI in agriculture through sensor-based smart cultivation practices, precision farming, soil mapping and smart breeding techniques, to name a few, has helped to track the current status of the crops and livestock, identify the associated risks and subsequently bring improvements by countering the negative impacts. Weather based sensors help to continuously monitor the weather parameters like temperature, humidity and other factors that might cause irreversible damage to the crops. Agriculture 4.0 technologies offer the farmers a more organized and scientific approach towards farming with a high level of accuracy in final product estimation (Ferrag *et al.*, 2021, Price, 2022). Owing to its end-to-end visibility, IoT based technologies has made the farming sector more predictable and efficient (Javaid *et al.*, 2022), particularly in the face of rising consumer demand and climate change. Agriculture 4.0, thus, is on its way of transforming India from self-sustenance to self-reliance.

What is Artificial Intelligence?

The advent of Artificial Intelligence (AI) has brought a technological revolution in the agriculture sector. AI is a constellation of technologies that eases the problem-solving method by simply transferring the burden of decision-making to a processed algorithm. The three basic goals of AI include learning, reasoning and perception (Nawaz *et al.*, 2020)). AI has the ability to augment the human capabilities of sense, learn from experiences and adapt to changing situations over time. Thus, it enables machines to act with higher levels of intelligence. The concept of AI is not new. The term ‘Artificial Intelligence’ was coined by John McCarthy way back in 1955 (Bresnahan *et al.*, 1995), and since its inception about 70 years ago, it has been applied in many industries and even government sector, though in limited use. However, with increasing availability of large volume of data and decreasing costs of data storage, use of AI has undergone an exponential rise across various sectors in the recent years (Anon., 2018). However, the accomplishments of AI would not have been possible without the Internet of Things (IoT). The IoT refers to an interconnected network of technologies that have been designed to optimize the collection of massive amounts of data while maintaining effective communication between operations and minimizing human interaction. For instance, with the help of IoT, farmers are able to regularly monitor their cattle health in real-time through connected sensors, that spontaneously record the heart rate, blood pressure, body temperature of the animals. As such, farmers can be warned of any abnormal condition of the animal, thereby allowing for quick response and recovery (Javaid *et al.*, 2022). The advent of AI and IoT has made a major breakthrough across various sectors like health, education, agriculture, banking and financial services, infrastructural development, etc. In the agricultural sector, AI is seen to be a promising technology to drive a food revolution towards achieving the increased demand for food for a rising population.

Application of Artificial Intelligence in Agriculture

The agriculture sector, despite its notable advancements and government support, remains vulnerable to uncertain weather conditions, unstable supply chain, and experiences low productivity due to the following factors:

- Inefficient utilisation of resources
- Traditional cultivation practices which require a greater amount of time and labour
- Inadequate knowledge of the weather, soil, and fertilizer usage
- Traditional pesticide and herbicide spraying which harms farmers and lowers agricultural output
- Inadequate storage techniques which resulted in the deterioration of produce

The utilisation of AI in agriculture has fundamentally transformed conventional farming methods, converting them into advanced, data-centric procedures. AI technologies, including machine learning, computer vision, and robotics, are being used to improve different elements of agriculture, such as crop monitoring, management, predictive analytics, automated machinery, and resource optimisation. For instance, AI has the potential to diagnose pest incidence and disease infestation in real-time, and can offer localized advice to the farmers through mobile applications, thereby minimizing crop failures. Besides, AI contributes towards building climate

resilience by tracking the changing weather patterns and predicts how rural communities can be affected, thereby preventing the farming community from worst outcomes.

Crop Monitoring and Management

AI-enabled drones and satellite photography provide detailed photos of agricultural areas, which are then processed using AI algorithms to evaluate crop vitality, identify diseases, and detect insect invasions. By utilizing real-time data, farmers may make accurate predictions on the probability of disease outbreaks and insect invasions. These predictions are based on environmental circumstances and the growth phases of crops. The utilization of sensors has fundamentally transformed the process of soil and crop monitoring. These sensors gather data on soil moisture, nutrient levels, temperature, and other environmental variables. This data is then examined by AI models to offer accurate suggestions for irrigation, fertilization, and other agricultural techniques. Through the analysis and correlation of data pertaining to weather patterns, seed varieties, soil conditions, pest infestations, disease occurrences, historical yields, market trends, prices, and customer demands, farmers can strategically determine the most profitable course of action to optimize crop profitability.

Enhancing the effectiveness of agricultural mechanisation

The integration of image classification techniques with remote and local sensed data has the potential to significantly transform the utilisation and effectiveness of farm machinery in various areas such as weed control, early disease detection, crop harvesting, and grading. AI-powered machinery, such as self-driving tractors and harvesters, carry out a range of agricultural chores with exceptional accuracy. These tools decrease labour expenses and improve operational effectiveness, enabling farmers to concentrate on other crucial tasks. AI-driven robots are employed for the purpose of weed management, utilising advanced technology to detect and eradicate unwanted plants while minimising the need for chemicals. This not only diminishes the utilisation of chemicals but also encourages the adoption of sustainable farming methods.

Resource Optimisation

It refers to the process of maximising the efficiency and effectiveness of available resources. AI systems enhance water use efficiency by analysing data on soil moisture and weather forecasts. This practice ensures that crops are provided with the appropriate amount of water at the correct timing, thereby conserving water resources and enhancing crop health. The utilisation of image recognition and deep learning models has facilitated the dispersed monitoring of soil health, eliminating the requirement for laboratory testing equipment. By combining AI technology with data from satellites and on-site image capture, farmers can now promptly address soil health issues. This is achieved by receiving recommendations for the most suitable fertiliser application, taking into account soil nutrient levels and crop needs. This focused strategy decreases the amount of fertiliser that is wasted and lessens the negative effects on the environment.

Supply Chain Management

The current inadequate levels of price realisation for farmers (as low as 20% in fruits and vegetables) can be attributed mostly to poor methods for determining and communicating prices,

inefficiencies in supply chain intermediaries, and local regulations. Utilising AI techniques for predictive analytics can provide farmers with more precise supply and demand data, hence diminishing the information asymmetry that exists between farmers and intermediaries. Given the global interconnection of commodities markets, the utilisation of big data analysis becomes essential. The data obtained from e-NAM, Agricultural Census (including information on more than 138 million operating holdings), AGMARKET, and over 110 million Soil Health Samples offer the necessary quantities for any predictive modelling (Anon., 2018). AI aids in forecasting inventory requirements and market demand, facilitating enhanced management of the supply chain. This minimizes waste and guarantees prompt delivery of produce to the market. AI algorithms evaluate the quality of harvested crops, ensuring that only the highest quality supply is delivered to consumers. This is especially crucial for export markets that have strict quality requirements.

Advantages of AI in Agriculture

- 1. Enhanced Efficiency and Productivity:** AI technologies streamline diverse farming chores, diminishing labour expenses and enhancing operational efficiency. Autonomous machinery and robots have the capability to carry out accurate farming tasks, hence improving production.
- 2. Advanced Crop Monitoring:** Drones, satellite images and IoT devices offer up-to-date information on the condition of crops, soil, and environmental elements. This enables the fast identification of diseases and pests, prompt interventions, and improved resource management.
- 3. Enhanced Decision-Making:** Predictive analytics and forecasting models assist farmers in making well-informed decisions pertaining to crop planning, yield prediction, and market strategy. As a result, this leads to the efficient distribution of resources and a decrease in potential hazards.
- 4. Sustainable Farming Practices:** AI systems facilitate precision farming, which lowers the need for pesticides, fertilisers, and water. This initiative encourages the use of sustainable farming methods, reduces the negative effects on the environment, and preserves valuable natural resources.
- 5. Supply Chain Optimisation:** AI enhances supply chain optimisation by facilitating inventory and demand forecasting, hence ensuring the effective management of the supply chain.

Quality control systems provide stringent standards for agricultural produce, enhancing marketability and ensuring consumer pleasure.

Disadvantages of AI

- 1. Substantial Initial Capital Requirement:** The integration of AI technology necessitates a considerable upfront expenditure on hardware, software, and infrastructure. This can provide a hindrance for small-scale and economically disadvantaged farmers, as well as farmers with limited resources.

2. Insufficient Proficiency: The operation and maintenance of AI systems necessitate a level of technical knowledge and competence. Adequate training and support may be required by farmers to proficiently utilise AI tools and accurately interpret data.

3. Insufficient Awareness: Farmers possess inadequate knowledge regarding these technologies and therefore tend to favour traditional approaches.

4. Privacy and data security: The fundamental obstacle lies in the gathering and handling of substantial amounts of data that are crucial for AI algorithms, with regards to data privacy and security. This encompasses meteorological data, soil characteristics, crop vitality, and additional factors. It is imperative to guarantee the safeguarding of sensitive data.

5. Dependency on Accuracy and Data Quality: For AI models to work well, accurate and high-quality data are necessary. Erroneous or insufficient data can result in negative agricultural decisions.

6. Issues related to maintenance and operations: Regular maintenance and calibration are necessary for AI-powered machines and gadgets. Malfunctions or periods of inactivity have the potential to interrupt agricultural activities and impact overall productivity.

Role of Stakeholders and Government Initiatives

The integration of AI in agriculture is propelled not only by technology progress but also greatly affected by diverse government efforts and the participation of several stakeholders. These entities are essential for creating a favourable environment for the integration of AI technologies in farming practices.

Government Interventions in the form of Policies and Subsidies

The Government of India has launched many comprehensive national AI programmes that incorporate explicit provisions to enhance productivity and sustainability of the agricultural industry. Governments offer financial incentives, such as subsidies, grants, and low-interest loans, to incentivize farmers to embrace AI technologies. These financial subsidies assist in mitigating the substantial upfront expenses linked to AI tools and technology. To make Indian agriculture future-ready, the government has already taken initiatives like National Mission for Sustainable Agriculture, promotion of scientific warehousing and the adoption of drone technologies (Anon., 2023).

Government Initiatives for Research and Development

Government entities allocate funds to agricultural research institutes with the purpose of developing and evaluating AI applications that are specifically designed to suit the unique farming conditions of the local area. These institutes form partnerships with universities, technology businesses, and international organisations to promote the progress of AI research in the field of agriculture. Public-private partnerships (PPPs) are formed to harness the respective advantages of both the public and private sectors.

Development of AI Infrastructure

The Government has allocated resources to improve digital infrastructure, such as extending internet access in rural regions. Dependable internet connectivity is essential for farmers to make use of AI-driven products and obtain real time data for weather forecasts, soil health, market trends, and other crucial information. In India, Centre for Development of

Advanced Computing (C-DAC) has established AI Research Analytics and Knowledge Dissemination Platform (AIRAWAT) at Pune under the initiative of Ministry of Electronics and IT, Government of India, which is a high-performance supercomputing facility of 200 AI Petaflops. It can perform tasks like image recognition, speech recognition, natural language processing for research and for the support of advancements in the field of Agriculture (Anon., 2018).

Role of Academic Institutes and Research Centres

Academic institutions engage in state-of-the-art research on the implementation of AI in the field of agriculture. Their work involves the development of novel algorithms, the creation of AI models, and the rigorous testing of these technologies in authentic agricultural environments. Research organisations and universities provide training programmes to educate farmers, agronomists, and agricultural scientists on the utilisation of AI technologies. They have a crucial role in developing the necessary technical skills to use AI in agriculture.

Role of Private Enterprises

Private sector enterprises are leading the way in the development of AI technology for the agricultural sector. They develop AI-driven machinery, software solutions, and digital platforms to meet diverse agricultural requirements. These companies focus on the commercialization of AI technologies with the aim of providing access to farmers. In addition, they offer postpurchase support, which encompasses maintenance services and technical advice, to guarantee the optimal utilisation of AI products. The FarmBot network is a good example of digital agriculture where smart agricultural robots are linked with AI and IoT to process information autonomously and help farmers to take remedial action (Tao *et al.*, 2021, Liu *et al.*, 2021).

Role of Agricultural Associations and Cooperatives

Farmers' organisations and cooperatives promote the implementation of AI technologies among their members. They conduct workshops, demonstrations, and awareness campaigns to educate farmers about the advantages and practical uses of AI. These organisations also participate in collective bargaining with technology providers and government agencies to get advantageous terms and conditions for their members, secure financial assistance and reductions in prices.

Role of Non-Governmental Organisations (NGOs)

Non-governmental organisations (NGOs) are essential in enhancing the ability of smallholder farmers to embrace AI technologies. The organisation provides training programmes, disseminates educational resources, and offers practical assistance to farmers. NGOs frequently launch pilot initiatives to showcase the efficacy of AI in the field of agriculture. They compile and record accounts of successful experiences and detailed analyses of specific cases, which can be utilised to promote the broader acceptance and utilisation of these technologies.

Conclusion:

Since the dawn of agriculture, information on weather condition, soil health, nutrient quality and quantity, irrigation requirement and their projections have been very crucial for the

farming community. As agriculture has evolved with the touch of modernization, in response to increased production, efficiency, and fewer emissions, information must be more real-time, precise, and dependable. AI has the potential to be a game-changing technology which could help the farmers to decide on the exact requirement of an input and its exact time of application, which would also contribute towards lesser carbon emissions. Farmers would be able to take decisions based on previous experiences and learning. Thus, through the application of AI, sustainable decisions could be taken in the agriculture sector.

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DRONES IN INDIAN AGRICULTURE: TRANSFORMING FARMING PRACTICES FOR A SUSTAINABLE FUTURE

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Abstract:

The integration of drone technology into agriculture represents a pivotal advancement in modern farming practices. This chapter examines the progression of drones in agriculture, from their initial experimental uses to their current role as essential tools in the farming landscape. The discussion traces the development of drone technology, highlighting significant milestones and innovations that have enhanced their ability to address the diverse challenges faced by the agriculture sector. The chapter explores the varied applications of drones in agriculture, illustrating how these unmanned aerial vehicles are transforming crop monitoring, precision agriculture, pest control, and soil analysis. It emphasizes the tangible impact of drones in revolutionizing traditional farming methods, providing insights into increased efficiency, cost reduction, and improved crop yields. Additionally, the chapter highlights the benefits of drone technology in agriculture, demonstrating how these aerial devices enhance overall efficiency, contribute to cost savings, and optimize crop production. The environmental benefits, such as reduced pesticide usage and water conservation, are also examined, positioning drone technology as a key component of sustainable farming practices.

Keywords: Drones, Indian Agriculture, Precision Farming, Remote Sensing, Pest Surveillance, Crop Monitoring, Yield Estimation.

Introduction:

Agriculture, as the world's primary source of food, faces significant challenges including rising demand, food safety concerns, and the need for environmental sustainability (Friha *et al.*, 2021). With the global population anticipated to reach 9.7 billion by 2050, the demand for food and water is expected to increase substantially. Issues such as limited arable land, a declining number of farmers, and environmental concerns highlight the necessity for innovative and sustainable farming solutions (Elijah *et al.*, 2018). To tackle these challenges, the integration of advanced technologies like smart farming and precision agriculture has gained considerable attention. Smart farming involves the use of cutting-edge innovations, including information communication technologies, to improve farming efficiency (Haque *et al.*, 2021). Precision agriculture emphasizes site-specific management using technologies such as Wireless Sensor Networks, the Internet of Things, artificial intelligence, computing technologies, big data, and blockchain (Pincheira *et al.*, 2021).

Remote sensing technologies, including satellites, manned aircraft, and drones (Unmanned Aerial Vehicles or UAVs), are essential for advancing smart and precision agriculture. Drones, in particular, offer several benefits such as high-quality imaging on cloudy days, cost-effectiveness, ease of setup and maintenance, and swift data transfer. Initially developed for military use, drones have now found applications in various civilian sectors, including agriculture. They are useful for crop management, disaster reduction, early warning systems, and conservation efforts in wildlife and forestry (Huang *et al.*, 2021). In the agricultural sector, drones are instrumental in activities like crop monitoring, yield estimation, water stress assessment, and the detection of weeds, pests, and diseases (Inoue, 2020). Furthermore, drones facilitate precision irrigation and the management of weeds, pests, and diseases based on environmental data.

Overview of Agriculture Challenges:

Precision Farming:

Precision farming, also referred to as precision agriculture, is a fundamental challenge in modern agriculture. Traditional farming methods are becoming insufficient to meet the needs of a growing global population. The variability in soil types, weather conditions, and crop requirements calls for a more precise and targeted approach. Precision farming seeks to optimize inputs like water, fertilizers, and pesticides by adjusting them to the specific needs of different areas within a field. The main challenge is to develop methods that can achieve this level of precision in an efficient and cost-effective manner.

Crop Monitoring:

Accurate and timely crop monitoring is a significant challenge in agriculture. Traditional manual monitoring methods are labor-intensive, time-consuming, and often imprecise. Early detection of diseases, nutrient deficiencies, or pest infestations is crucial for preventing yield losses. Additionally, monitoring crop health helps optimize irrigation and fertilization practices. As farming operations expand, the need for solutions that offer comprehensive, real-time data on crop conditions becomes even more critical.

Resource Optimization:

Optimizing resources such as water, energy, and land remains a constant challenge in agriculture. With global water scarcity on the rise, innovative irrigation methods are crucial. Energy-intensive farming practices lead to environmental degradation, making energy efficiency a priority. Efficient land use is vital to balance increased food production with the preservation of natural ecosystems. The goal is to develop sustainable practices that enhance resource efficiency while maintaining or improving crop yields and quality.

Climate Variability:

Climate change adds another layer of complexity to agricultural practices. Unpredictable weather patterns, extreme events, and shifting climatic zones challenge farmers who traditionally depend on historical weather data for decision-making. Adapting to these changes necessitates dynamic and responsive strategies that can adjust to evolving environmental conditions.

Why Undertake Agricultural Drones?

Drone technology has gained significant popularity in the agricultural industry due to its versatility and is seen as the future for the farming community. Initially used by the military, drones now enhance overall performance and help farmers address various challenges, offering numerous benefits through precision agriculture. Agricultural drones help bridge the gap caused by human error and inefficiency associated with traditional farming techniques. The aim of adopting drone technology is to eliminate guesswork and provide accurate and reliable data. External factors such as climate, soil conditions, and temperature are crucial in farming. Agricultural drones enable farmers to adapt to specific environments and make informed decisions accordingly. The data collected by drones' aids in adjusting crop health, treatment, scouting, and irrigation, as well as conducting soil analysis and assessing crop damage. Drone surveys help increase crop yields while reducing time and costs.

How Does Drone Generation Work?

A comprehensive understanding of drone features is essential to fully appreciate the benefits of agricultural drones. Typically, drones are equipped with a navigation system, GPS, multiple sensors, cameras, programmable controllers, and tools for autonomous operation. Currently, many farmers use satellite imagery as an initial guide for farm management. However, equipped with advanced technology, unmanned aerial vehicles (UAVs) can capture more precise data than satellites for precision agriculture. This data is then processed through agri-tech software to generate valuable insights. The data collection from agricultural drones typically occurs in the following stages:

- **Reading the location:** This identifies the territory being tested. Therefore, the first step consists of establishing a boundary, analyses of the place, and then finally, importing the technical GPS data into the drone's navigation machine.
- **The usage of self-sustaining Drones:** In view that unmanned aerial motors (UAVs) are unbiased, they input flight patterns into their already installed machine to collect required information.
- **Importing the information:** After capturing all the required data through sensors consisting of the multispectral sensor/RGB sensor and it became processed through numerous software for addition analysis and interpretation.
- **Output:** After gathering the records, they format it so that farmers can recognize the information and not using a trouble, bringing them a step towards precision farming. 3-D mapping or Photogrammetry are popular techniques to show enormous facts accumulated.

Role of Drone Technology:

In addressing these challenges, technology stands as a beacon of hope for the agriculture sector. The integration of advanced technologies, including drones, sensors, and data analytics, promises to revolutionize traditional farming practices. By harnessing the power of data, farmers

can make informed decisions that optimize their operations for increased efficiency and productivity. Technology-driven solutions in agriculture not only tackle existing challenges but also pave the way for sustainable and resilient farming practices. Drones, in particular, provide a unique perspective, allowing farmers to monitor large areas of land with exceptional detail. The ability to gather real-time data on crop health, soil conditions, and overall farm performance enables farmers to respond proactively to issues, thereby enhancing the sustainability and productivity of agriculture. As we explore the applications of drone technology in agriculture, it becomes clear that these aerial systems are more than mere tools; they are catalysts for a fundamental shift towards precision, efficiency, and sustainability in farming practices.

Types of Drones Used in Agriculture:

- **Fixed-Wing Drones:** Fixed-wing drones, characterized by their airplane-like design, thus covering large agricultural areas efficiently. Equipped with long-range capabilities and stable flight patterns, these drones are well-suited for mapping expansive fields, monitoring crop health, and assessing overall farm conditions.
- **Rotary-Wing Drones (Quadcopters and Hexacopters):** Rotary-wing drones, often in the form of quadcopters or hexacopters, provide agility and versatility. Their ability to hover, take off vertically, and navigate confined spaces make them ideal for close-up inspections, detailed imaging, and precise data collection in smaller or irregularly shaped fields.
- **Hybrid Drones** Hybrid drones combine features of both fixed-wing and rotary-wing designs, offering a balance between endurance and flexibility. These drones can cover large areas efficiently and then transition to rotary-wing flight for detailed inspections or data collection in specific regions of interest.

Applications of Drones practices in Agriculture:

Drone technology speedy reestablishes traditional agrarian practices and is subsequently accomplishing them as follows; Irrigation monitoring, crop fitness, crop harm assessment, discipline soil evaluation, precision agriculture, faraway sensing, synthetic intelligence (Dutta and goswami, 2020)

❖ **Irrigation tracking**

Drones equipped with hyperspectral, thermal, or multispectral sensors can identify areas that are excessively dry or in need of improvement by farmers. Drone surveys help enhance water efficiency and identify potential pooling or leaks in irrigation systems by providing irrigation monitoring and yield calculations based on plant index to assess crop health and detect emitted heat or energy.

❖ **Crop health tracking and Surveillance**

It is essential to monitor plant health and detect bacterial/fungal infestations in the early stages. Agricultural drones can detect variations in the reflection of green light and near-infrared spectroscopy (NIRS) light by different plants. This data helps generate multispectral images to monitor crop health. Timely monitoring and detection of any abnormalities can help prevent crop

loss. In the event of crop failure, farmers can also provide accurate documentation for insurance claims.

❖ **Water strain monitoring**

Assessing water stress on plants is a complex task due to the multifaceted effects of drought, which can be influenced by various factors (Espinoza *et al.*, 2017). Variables derived from thermal images often rely on subtle temperature variations to detect stress and other phenomena. Consequently, thresholds and regression equations developed under specific conditions may not hold true under slightly different circumstances. For example, different genotypes of the same crop may exhibit significantly different canopy temperatures under identical conditions due to inherent variations in stomatal conductance and transpiration rates.

❖ **Crop harm evaluation**

Agricultural drones equipped with multispectral sensors and RGB sensors can identify areas in fields affected by weeds, infections, and pests. With this information, precise amounts of chemicals needed to combat these infestations can be determined, helping to reduce costs for farmers.

❖ **Nutrient status and deficiency tracking**

Plants require an optimal balance of nutrients to ensure healthy growth and high yield. Nitrogen is crucial for vigorous plant growth and foliage development, while phosphorus is essential for strong root and stem formation. Potassium is vital for enhancing disease resistance and improving crop quality. If any of these nutrients are deficient in the soil, plants can become stressed and struggle to thrive. Laboratory leaf analyses can provide accurate evaluations of nutrient levels, but these are time-consuming and require specialized techniques for precise interpretation. Indirect methods, such as using a chlorophyll meter (SPAD) for estimating nitrogen levels, are available. However, this approach is also time-consuming and can sometimes yield inaccurate estimates. Therefore, significant efforts have been made to develop new methods for detecting and estimating nutritional issues in plants. The development of advanced techniques aims to provide quicker and more accurate assessments of plant nutrient status, helping farmers to manage soil fertility more effectively and ensure optimal plant health and productivity.

❖ **Field soil analysis**

Drone surveys enable farmers to gather essential information about soil conditions on their land. By utilizing multispectral sensors, drones can capture data useful for optimizing seed planting patterns, conducting comprehensive soil analysis, managing irrigation, and monitoring nitrogen levels. Additionally, photogrammetry and 3D mapping technologies allow farmers to thoroughly assess their soil conditions, facilitating better decision-making for crop management.

❖ **Diseases tracking**

Crop diseases can be devastating and are typically categorized as fungal, bacterial, or viral. Drones equipped with infrared cameras can detect issues within plants (Klemas 2015), providing a clear picture of their health. If a farmer can identify an infection early, preventative

measures—such as removing the infected plant—can be taken to prevent the spread to neighbouring plants. Image-based tools are therefore crucial in detecting and diagnosing plant diseases, especially when human assessment is inadequate, unreliable, or unavailable, particularly given the extensive coverage provided by UAVs. While RGB and multispectral images are commonly used for gathering data on affected areas, hyperspectral and thermal images have also been tested. Thermal imaging, in particular, is used to detect signs of water stress, which may be indicative of disease (Dash *et al.* 2018).

❖ **Weed control**

Weeds are undesirable plants that grow among agricultural crops and can cause significant problems. They compete for resources like water and space, leading to reduced crop yields. In India, yield losses due to weeds are substantial, affecting various crops: Rice (10-100%), Wheat (10-60%), Maize (30-40%), Sugarcane (25-50%), Vegetables (30-40%), Jute (30-70%), and Potato (20-30%). Traditionally, herbicides are the primary method for weed control. Farmers often uproot weeds post-emergence and commonly spray herbicides uniformly across entire fields, even in weed-free areas. However, excessive use of herbicides can lead to herbicide-resistant weeds and negatively impact crop growth and yields. To address these issues, advanced methods using hyperspectral and RGB imaging are being explored. Hyperspectral images can discriminate between the spectral signatures of weeds with different resistances to herbicides like glyphosate. RGB sensors can classify various weed species. Researchers have also used drones equipped with hyperspectral sensors to monitor weeds based on plant canopy chlorophyll content and leaf density (Malenovsky *et al.*, 2017).

These advanced imaging techniques offer a more precise approach to weed management. By generating accurate weed cover maps, drones can identify specific areas where herbicides are needed, reducing overuse and minimizing environmental pollution. This site-specific weed management strategy ensures targeted herbicide application, effectively controlling weeds while conserving resources and protecting the environment.

❖ **Planting**

In India, drone startups have developed drone-planting systems that enable drones to deploy pods containing seeds and essential nutrients directly into the soil. This innovative technology significantly reduces costs by up to 85% while enhancing consistency and efficiency in planting operations.

❖ **Drone for Evapotranspiration (ET) estimation**

Evapotranspiration (ET) is a critical process in which water is transferred from land to the atmosphere through evaporation from the soil and transpiration from plants. Estimating potential ET is crucial for experts in hydrology, agriculture, and water management, particularly given the challenges of water scarcity, growing populations, and climate change. Unmanned aerial vehicles (UAVs) are increasingly used for ET estimation in various research contexts. There are primarily three types of UAV systems used: aircraft, fixed-wing, and quadcopter. Aircraft UAVs are typically more expensive but can fly longer and carry heavier sensors. In contrast, fixed-wing

and quadcopter UAVs are more affordable. Fixed-wing UAVs can generally fly for about two hours, making them suitable for large-scale field studies. Quadcopters, with a flight time of approximately 30 minutes, are ideal for short missions in smaller fields. Using UAVs as remote sensing platforms introduces new research opportunities and challenges, such as drone image processing and flight path planning. For instance, fixed-wing UAVs can collect thermal data to estimate ET using two-source energy balance models. These capabilities enhance the precision and efficiency of ET estimation, supporting better water resource management and agricultural planning.

❖ **Insecticides spraying**

Using drones for crop spraying significantly reduces human exposure to hazardous chemicals. Agri-drones can perform this task much more quickly than traditional methods such as tractors or airplanes. Equipped with RGB and multispectral sensors, drones can accurately identify and treat problematic areas in fields. Experts note that aerial spraying with drones is up to five times faster compared to other methods.

❖ **Farm animals tracking**

The implementation of drone surveys has revolutionized farming practices by not only facilitating crop monitoring but also by enabling the tracking of livestock movements. With the aid of thermal sensor technology, drones can efficiently locate lost animals and identify instances of injury or illness. This functionality significantly contributes to overall crop yield. The utilization of drones in agriculture spans a wide array of applications, providing farmers with a diverse set of tools to enhance operational efficiency, productivity, and environmental sustainability. In the following section, we will delve into the various applications of drones in contemporary agriculture, including precision farming, pest control, and soil analysis.

Benefits of Drone Technology in Agriculture:

Drones provide substantial advantages in modern agriculture, revolutionizing traditional farming practices. Some key benefits include:

❖ **Crop Monitoring and Management:**

Drones equipped with high-resolution cameras and sensors enable comprehensive crop monitoring. This technology empowers farmers to closely observe the health, growth, and overall condition of their crops with unprecedented precision. By harnessing real-time data obtained from drone imagery, farmers can make proactive decisions to address potential issues such as nutrient deficiencies, diseases, or pest infestations.

❖ **Yield Estimation:**

Drones play a crucial role in providing accurate yield estimation by analysing both crop density and health, as discussed by Panday *et al.* (2020). This valuable information assists farmers in effectively planning harvest schedules and optimizing storage and transportation logistics.

❖ **Water Stress Assessment:**

Drones equipped with either thermal or multispectral sensors have the capability to identify areas within crops experiencing water stress. This enables farmers to implement precise irrigation strategies, conserving water resources and enhancing overall water-use efficiency.

❖ **Weed, Pest, and Disease Detection:**

Drones play a pivotal role in the early detection of weeds, pests, and diseases. Gasparovic *et al.* (2020) note that high-resolution images captured by drones enable the identification of specific areas affected, facilitating precise and timely application of pesticides or herbicides. This targeted approach minimizes the necessity for widespread chemical use, promoting more sustainable farming practices.

❖ **Precision Agriculture:** Drones contribute to the principles of precision agriculture by offering intricate spatial data, as outlined by Kalischuk *et al.* (2019). This capability enables the customization of farming practices on a site-specific level, thereby optimizing the utilization of resources such as water, fertilizers, and pesticides.

❖ **Cost-Effectiveness:**

When contrasted with traditional aerial surveys or manual labour, drones present a cost-effective approach to data collection. They are relatively straightforward to deploy and capable of covering extensive agricultural areas swiftly, thereby diminishing labour costs and boosting operational efficiency.

❖ **Improved resolutions in both temporal and spatial sensing**

Improving the temporal and spatial sensing resolutions of drones holds paramount importance for enhancing data capture over time and increasing the detail in images. This technological progress enables more frequent monitoring of changing conditions and delivers clearer, more detailed images of agricultural landscapes. Emphasize that the combined impact of enhanced temporal and spatial resolutions renders drone's indispensable for precision agriculture, facilitating accurate and timely decision-making across various applications.

Challenges and Limitations of Drone Technology in Agriculture:

The integration of drones into agriculture offers manifold advantages, yet it's crucial to recognize and tackle the obstacles and constraints accompanying this revolutionary technology. In this segment, we explore the primary challenges encountered by farmers and stakeholders in embracing and harnessing the full potential of drone technology in agriculture."

Regulatory Challenges:

A significant barrier to the widespread adoption of drone technology in agriculture is the intricate and ever-changing regulatory environment. Rules and regulations differ significantly from one region to another, affecting the permissions, licensing, and operational limitations imposed on drone usage. Farmers frequently face challenges in complying with these regulations and navigating bureaucratic procedures, which can hinder the smooth incorporation of drones into everyday farming activities (Ayamga *et al.*, 2021).

Cost Barriers:

Despite the decreasing costs associated with drone technology, the initial investment still presents a formidable barrier for numerous farmers, particularly those operating on smaller scales. Procuring high-quality drones outfitted with advanced sensors and imaging technologies, alongside acquiring requisite software and training, can pose a financial obstacle. Moreover, ongoing maintenance, insurance, and potential upgrades add to the overall expense of adopting drones in agriculture.

Conclusion:

Drone technology represents far more than just a technological novelty; it signifies a paradigm shift in agricultural practices. Drones' capacity to offer actionable insights, refine precision, and tackle longstanding challenges positions them as indispensable tools for contemporary farmers. Their benefits in terms of efficiency, cost reduction, and sustainability underscore their pivotal role in shaping the future of agriculture. To fully unleash the potential of drone technology in agriculture, sustained research, investment, and widespread adoption are essential. Collaboration among researchers, technologists, policymakers, and agricultural stakeholders is imperative to address current challenges, foster innovative solutions, and streamline regulatory frameworks. Investment in educational programs is vital to ensure that farmers, irrespective of their scale or location, possess the knowledge and skills necessary to harness the advantages of drone technology. As we envision the future of agriculture, drones emerge at the forefront of a technological revolution capable of enhancing productivity, fostering sustainability, and ensuring food security for an expanding global population. By embracing and investing in drone technology, we lay the groundwork for a more resilient, efficient, and environmentally conscious agricultural sector.

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INTEGRATED PEST MANAGEMENT STRATEGY FOR THE MANAGEMENT OF POMEGRANATE, *PUNICA GRANATUM* (LINN.) INSECT PESTS

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Abstract:

Pomegranate (*Punica granatum* L.), a fruit of important nutritional and economic value, faces major contests in commercial production due to various insect pests. It includes the Pomegranate Fruit Borer, Aphids, Whitefly, Thrips and Mealybugs provide to substantial yield and quality losses. Integrated pest management system corresponds of using suitable ways and styles in a compatible manner to maintain pest populations at situations below those causing economically inferior damage or loss. IPM combines cultural control, biological control and chemical measures to give a cost-effective, environmentally sound and socially respectable system of controlling conditions, insects, weeds and other. It involves the pre-season operation of pests through guidance in selection of crops and cultivars suited to particular soils, timely planting, nonstop monitoring of crop health and pest status, conservation practices for native natural adversaries, and the use of timely and quality inputs of bio-rational integrated with position-specific crop product practices.

Keywords: Pomegranate, Insect pest, Borers, Management strategies

Introduction:

Pomegranate, *Punica granatum* (L.), is a fruit-bearing shrub or small tree that is grown in most parts of the world, mainly in arid and semi-arid region. Pomegranate, a fruit known for its sweet and sour taste and multiple health benefits have now become popular and widely cultivated, mostly grown in places such as India, Iran, the United States, and Spain. Nevertheless, industrial cultivation of pomegranate is stifled with several constraints, among which pest management particularly insect pests is the major threat. In recent years, integrated insect pest management programmes have become necessary to ensure a high yield and quality of pomegranate fruits in order to keep pomegranate farming economically sustainable. Around 45 species (Butani, 1979) 32 insect and non-insect pests (Balikai, 2000), more than 50 species of insects (Verghese and Jayanthi, 2001) and 33 insect pests (Balikai *et al.* 2003) cause damage in pomegranate plant. In India, there are 91 insects, 6 mites, and 1 snail pest that feed on the pomegranate crop. By drying, biting, and boring blossoms, fruits, twigs, and foliage, pomegranate pests cause significant damage which reduces fruit quality and decreases yield. In recent times, the cultivation of high-yielding pomegranate types under intense care and management, under irrigated conditions, with early plant exploitation, has resulted in some

serious insect issues. Among them, infestation by sucking pests like aphids, thrips, whiteflies, mealy bugs, scale insects and mites results in reduction of pomegranate fruit yield and quality.

Major Insect Pests in Pomegranate

Many insect pests can adverse effect on pomegranate crops, they reduced yield and also fruit quality. These five major significant insect pests includes here:

1. Pomegranate Fruit Borer, *Deudorix isocrates* (Lepidoptera: Lycaenidae)

Fruit and seeds are affected by the larvae of these pest. Caterpillar bores into young fruits feeds on internal contents (pulp and seeds). Infested fruits have holes with blackish brown excreta of larva on surface. The fruit borer infected fruits attract bacteria and fungi which cause rotting in the fruits producing foul smell.

2. Pomegranate Aphid, *Aphis punicae* (Homoptera: Aphididae)

It infests new flush of pomegranate. They feed by sucking the cell sap from leaves and tender twigs. The affected parts get discoloured and disfigured. These insects also secrete a copious amount of honeydew on fruit, rot and shooty mold may appear. A severe infestation causes yellowing of leaves, wilting of terminal shoots or stunting of the tree.

3. Whitefly, *Siphoninus phillyreae* (Hemiptera: Aleyrodidae)

Flying insects causes yellowing, withering, and eventually drop of leaves by sucking sap from the vegetation. Serious damage is caused by the excretion of honeydew secreted by whitefly. Under moist conditions, shooty moulds can appear on honeydew.

4. Mealybugs, *Planococcus citri* (Hemiptera: Pseudococcidae)

Whole plant is infected by the bug. Nymphs and adults alike sip the sap off leaves, flowers, and fruits, causing the leaves to turn yellow, the flowers to curl and shed, and the fragile fruits to fall off. These pests reduce the market value of fruits. During dry season insects can get into roots and suck the sap. Sooty mold may appear on leaves and fruits (Sunil *et al.* 2023).

5. Thrips, *Scirtothrips dorsalis* (Thysanoptera: Thripidae)

Which is often seen on leaves and also on young fruits causing characteristic scab on fruits and thereby reducing the market and export value. When severe on leaves, it causes leaf tip curl and drying and shedding of flowers. The yield is drastically reduced. Feeding on new growth of plants. Nymphs and adults lacerate and suck the sap from buds, flowers.

Integrated Pest Management (IPM) Strategies

Integrated pest management has been enshrined as cardinal principle of Plant protection in the overall crop protection programme. IPM is an eco-friendly approach for managing pest problems encompassing available methods and techniques of pest control such as cultural, biological, mechanical and chemical control. It is an ecosystem approach to crop production and protection. It incorporates several management techniques and methods (Dubey *et al.* 2013).

1. Cultural Practices

Gather and dispose of agricultural waste. When the crop is nearing its crucial stages, provide irrigation. Steer clear of water logging Prevent water stress when the plant is blooming.

Make prudent use of fertilizers and avoid using chemical pesticides when 1-2 larval parasitoids are seen in the agricultural field to increase parasite activity (Das *et al.* 2024).

2. Biological Control

The biological control is the most important practice of integrated pest management, or IPM. In general, biocontrol refers to the employment of living organisms such as pests to reduce other living species. Stated differently, the intentional utilization of parasitoids, predators, and diseases to control the population of pests at a level that does not cause economic loss, either by introducing a new bio agent into the pest's habitat or by enhancing the efficacy of those already present in the field. Some parasitoids and predators are these:

Parasitoids: *Trichogramma*, *Ceranisus menes*, *Encarsia inaron* (Nymph parasitoid), *Tetrastichus spp.*, *Telenomus spp.*, *Chelonus blackburni*, *Braconid wasp* (Larval parasitoids) *etc.* Predators: *Menochillus sexmaculatus*, *Rodolia fumida*, *Ground beetles*, *Rove beetles*, *Spiders*, *Predatory mite*, *Pentatomid bug etc.*

3. Mechanical Control

Used of Traps and Barriers for control pests like pheromone traps, sticky traps and physical barriers can help the minimize pest population. Manual removal of pests in small orchards. It is effective method to manage infestations.

4. Chemical Control

Pesticides spray should be less toxic to natural enemies with low residual toxically to pollinators; safe to mammals and human being. Using selective insecticides that target specific pests and also used recommended application rates and timing to evade pesticide resistance and environmental contamination. Pesticides spray should be less toxic to natural enemies with low residual toxically to pollinators are safe to mammals and human being.

1. Spray Dimethoate 0.06% or 0.5% multineem, is useful to control thrips and aphids infestation.
2. To take up one spray of *lambda cyhalothrin (0.0025%) in evening hours, to recede the per-centage damage by whitefly.
3. Spraying with decamethrin at 0.0028% at the time when more than 50% of fruits have set (Anon., 2000). Repeat after two weeks with carbaryl at 0.2% or fenvalerate at 0.005%. In non-rainy season quinalphos at 0.06% was effective (Verghese and Jayanthi, 2001) for control pomegranate fruit borrar.
4. Spraying of insecticides like dichlorvos (0.02%) or malathion (0. 2%) with fish oil rosin soap was found to control the insect population. Application of phorate 10G (20 g/plant) is effective in controlling the pest population in the soil.

Monitoring and Surveillance

Regular monitoring for control pest populations and reduce their damage levels through field surveys. Farmers may make well-informed judgments regarding pest control tactics by putting decision support systems and pest forecasting models into place.

Conclusion:

Several insect pest management options are found in pomegranate cultivation. It requires a broad integrated method are cultural, biological, mechanical, and chemical control methods. By assuming IPM strategies, the growers of pomegranate can minimize pest damage. The farmer trust on chemical pesticides, and encourage sustainable farming practices. Continued research, education, and extension services are essential to support farmers in implementing effective pest management strategies and confirming the long-term productivity and profitability of pomegranate orchards.

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Farming the Future: Advanced Techniques in Modern Agriculture Volume II

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