



## Alien Gene Introgression Approach for Crop Improvement

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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.
- (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.
- (c) **Cross breeding:** Cross breeding is done to create hybrid plants that carry genetic material from both the crop and the donor species.
- (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 (F<sub>2</sub>, F<sub>3</sub>, etc.) to gradually increase the proportion of crop genes while retaining the desired alien genes.
- (e) **Selection and Marker-Assisted Breeding:** During backcrossing, marker-assisted selection is performed to choose plants that exhibit the target traits and have a higher proportion of crop genes.
- (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.
- (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.
- (i) **Release and Adoption:** Once a new crop variety is approved and deemed safe, it can be released to farmers for cultivation.

### Need for Alien Gene Transfer

- (1) The needs 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.
- (2) 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.

(3) Wild species are a rich reservoir of useful alien genes which are no longer available within the cultivated gene pool. 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

Here are some sources of alien genes and their characterization: (a) Wild Relatives of Crops (b) Other Cultivated Varieties (c) Cross-Compatible Species

### Characterization of Alien Genes

(a) **Phenotypic Characterization:** The first step in characterizing alien genes involves assessing the phenotypic traits associated with these genes.

(b) **Molecular Characterization:** Molecular techniques are used to identify and characterize the specific genes or DNA sequences responsible for the desirable traits.

(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.

(d) **Linkage Analysis:** Linkage analysis helps determine if the alien genes are tightly linked to other genes in the donor genome.

(e) **Functional Analysis:** Functional analysis involves determining how the alien genes function in the context of the target crop.

(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.

(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.

### 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.

### Importance of alien gene introgression

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.

(b) **Trait Improvement:** By incorporating genes from donor species, crops can acquire desirable traits that may not be present in their current genetic makeup.

(c) **Pest and Disease Resistance:** Alien gene introgression can confer resistance to specific pests and diseases that threaten crop production.

(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.

(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.

(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.

### **New genetic approaches for harnessing the natural variation**

(a) **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 parent with a homozygous genetic background of elite parent.

(b) **Advanced-backcross (AB-QTL) analysis:** (i) Candidate-gene association mapping (ii) Genome-wide association mapping, or genome scan

(c) **Multi-parent advanced generation inter-cross (MAGIC) population**

(d) **Next Generation Sequencing (NGS) Technologies**

(e) **TILLING**

(f) **Eco TILLING.**

### **Role of Molecular Markers in Alien Gene Introgression**

(1). **Marker-assisted selection and marker assisted backcrossing:**

(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:** 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.

(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 recurrent parent. Traditional approaches of recovery of RP genome take up to six back cross generations but the use of markers enables to achieve the same in even in second back cross.

(2) **Marker-assisted recurrent selection:** There are cases where quantitative variation is controlled by many genes (QTLs) with minor effect.

(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). These GEBVs are 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. 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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