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Application of Alien Gene in Crop Improvement

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Alien gene incorporated in crop plants has create great improvement in different crop species. Alien genes have contributed several traits in the crop plants which are not available in the cultivated background. Wild species are profuse resources of useful alien genes which are not available in the cultivated germplasm. These include genes for resistance to diseases and insect pests, for tolerance to drought, salinity, alkinity,temperature and other abiotic stresses as well as for quality traits. While most of the, alien gene incorporation in crop plants have been gotten through vertical gene transfer, horizontal gene transfer through transgenesis, somatic hybridization and, most timely, intragenesis and cisgenesis has invoked huge interest of the plant breeder globally. These techniques, lately helped by molecular markers and *in situ* hybridization, together have led to tranfered of hundreds of candidate gene in cultivated background of crop species, so improving their genetic potential. Alien gene detection and role of alien gene transfer in creating variability. Besides, the QTLs or genes or superior alleles for candidate gene identified through linkage mapping, association mapping, AB-QTL approach can be introgressed or pyramided in elite varieties or genotype of interest by using MAGIC, MABC, MARS or GWS approaches.

Introduction

20th century has witnessed huge 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 agronomy, 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 traditional hybridization, of late, helped by molecular marker technology and genetic transformation. The aim of all these techniques has been either exchange of genes between sympatric or neighbouring populations of crops and related taxa or transfer of genes from related taxa into the cultivated germplasm 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 artificial systems, thus it is faster and often mediated by hybridization followed by a number of back crossings and harsh phenotypic selections.

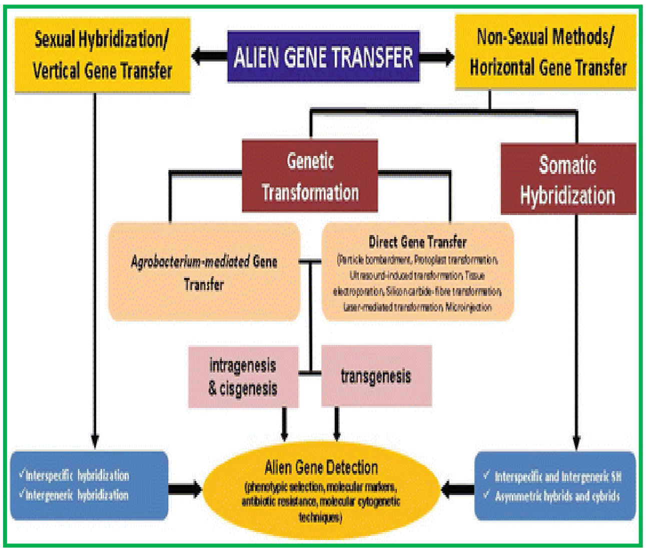
The introduction of foreign genes into the germplasm of a crop species by human intervention has been used by plant breeders and applied geneticists for almost 100 years. Natural selection and, of late, human selection have helped in maintaining new combinations of genes, and these gene combinations have been transferred through hybridization between cultivated and wild taxa leading to the development of populations with new characteristics and evolution of domestic crop. Breeders and geneticists have increasingly sought new sources of resistance in diverse germplasm, often involving distant and wild relatives (Gill *et al*. 2011). While there are several instances of wilfully introgression of desirable characters into crop cultivars as a part of regular plant breeding programmes, the extent and huge impact of farmer-aided or natural introgression are uncertain. During the crop domestication, few species of crops were selected for cultivation. As a result narrow germplasm forms the basis of modern monoculture in many areas of the world (Gill *et al*. 2011). So, use of vertical transfer for alien gene(s) could be restricted mainly to crossable wild species. Eventhogh, horizontal transfer of alien genes from non-crossable wild species or even across the genera is now increasingly being recognized as a significant and potent force in the evolution of eukaryotic genomes (Bock 2009).

While somatic hybridization after the initial leaps slowed down in yielding practical outputs, the capacity to transform crop plants has developed remarkably since the first genetically transformed plants were reported in 1983. Transgenics have a potential to hugely enhance the genetic component of integrated pest management through the development. The use of transgenics for crop protection from insects, disease and weeds is further expected to increase vigourly, while ethical and environmental concerns regarding the development and use of transgenic crop varieties may be addressed by latest introduction of technologies like intragenesis and cisgenesis.

Alien gene introgression is sometimes liked with several other obstacles such as linkage drag and pleiotropic effects; while in some instances this is liked with some unforeseen merits also such as development of chromosome elimination technique for doubled haploidy breeding in wheat and barley.

Alien Gene Transfer

Genetic variation is prerequisite for developing new plant varieties, and this can be created by introducing genes from a related wild 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. However, the valuable genes available within a crop species are easier to changes as compared to the alien genes, especially in the case of polygenic traits. Gene transfer within a species is not usually liked with undesirable effects which is one of the major demerit in alien gene transfer from distant species due to linkage of desirable genes with undesirable ones (linkage drag). In most of the cultivated crop species, limited popular and high yielding varieties are grown over wide range areas and these are often taken from a relatively narrow representation of germplasm, mostly from the primary germplasm, and so these have a narrow genetic base and limited genetic buffer. In this way, modern crop improvement although increased crop productivity worldwide, it also eroded the genetic variability of the crops (Hoisington *et al.* 1999). 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 gene pool forms the basis of modern monoculture in many areas of the world (Gill *et al*. 2011). This makes them tender to global climate change and prone to new races of pathogens and insect pests. Due to narrow genetic base, options to execute selection for desirable plant types also become limited. Since plant breeding in practice offers an option for plant breeding, efforts have been made to search for genes provides resistance to stresses within the cultivated species and to a limited extent among their wild relatives, but success has been limited to a few diseases and insect pests that are confined to monogenic gene from the primary germplasm in most of the crop plants (Kumar *et al.* 2011). To diversify and broaden the genetic base of cultivated germplasm, introgression of alien genes from wild species offers a viable option not only to reduced the risk of stress epidemics but also to make discernible yield advances in crop species. Wild species are a rich reservoir of useful alien genes which are no longer available within the cultivated germplasm.



Types of Alien Gene Transfer

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) (Fig. 1.1).

Factors Affecting Alien Gene Transfer through Hybridization

Hybridization is a frequent and significant component of plant evolution and speciation (Riesberg and Ellstrnd 1993). The value of hybridization in transfer of alien genes into cultivated crops has been known for a long time.

1. CHROMOSOME PAIRING: between chromosomes of the alien donor and those of the cultivated crops is go to gene transfers. For successful gene transfer through hybridization, cross-pollination must occur and there should not be any pre-crossing and post-crossing barrier. Necessary for hybridization include sympatric parents that occupy similar habitats, overlapping flowering times, like pollinators and intertaxa compatibility. Accordingly, gene transfer can be classified as intraspecific, interspecific and intergeneric. Eventhogh, the extent of gene transfer is affected by the objective of transfer, trait under transfer, breeding system of the partners involved, availability of pollinating agents, layout of the trial and population size. For the success of gene transfers between cultivated and wild species, chromosome pairing between chromosomes of the alien donor and cultivated species is most important as the success of gene transfer from donor to the recipient species depends upon the degree of chromosome homology that exists between these two species. Relatively easier gene transfers occur between diploid species such as maize and barley where only one genome needs to be constructed. 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 poly- ploid wheats and oat has resulted in desired chromosome pairing and hence alien gene transfers into these crop species (Jauhar 2006).

2. REPRODUCTIVE BARRIERS: Reproductive barriers limit alien gene transfer through interspecific and intergeneric hybridization. The sexual barriers stop distant hybridization have been distinguished into pre- and post-fertilization barriers (Stebbins 1958). These include those barriers that reduce the chances of formation of a viable zygote (pre-zygotic barriers) and those which are due to lower survival or reproductive fitness of the hybrids (post-zygotic barriers). The pre-zygotic barriers include ecological or habitat isolation, temporal differences in flowering phenology or pollinator service, temporal separation in flowering and/or pollination time between a pair of closely related species and gametophytic isolation. Gametophytic isolation is a pre-zygotic post-pollination mechanism which prevents fertilization by the pollen of foreign species.

3. HORIZONTAL GENE TRANSFER: Horizontal gene transfer (HGT), also called as lateral gene transfer (LGT), is a process in which a accepter organism take genetic material from a donor organism by asexual means (Bock 2009). Hence, it is the transfer of genes between the non-mating species and is not restricted by genome or germplasm boundaries. HGT is an adaptive force in avolution, contributing to metabolic, physiological and ecological innovation in most prokaryotes and some eukaryotes. HGT is primarily associated with prokaryotic species and it is expected to contribute up to 10–20 % of the genes in them (Nakamura *et al*. 2004) changes important characters such as photosynthesis, nitrogen fixation, virulence and antibiotic resistance. However, this phenomenon has also been found to occur in plant species on a huge scale . The most classical example of HGT in plants is the infection of plant cells with Agrobacterium. During infection, a region of the Ti plasmid (tumour-inducing plasmid) of the bacterium is incorporated in the nuclear genome of the plant cell. This phenomenon leads to development of modified Ti plasmids which are used as vehicles for introducing foreign genes into plants via Agrobacterium -mediated genetic transformation.

4. TRANSGENE INTROGRESSION

The infection of plant cells with Agrobacterium is a classic example of plants as recipients of HGT between kingdoms (Bock 2009). In case of transgenics the transferred gene usually derives from an alien species that is neither the recipient species nor a close, sexually compatible relative. The capicity to transform crop plants has developed hugely since the first transformed plants were reported in 1983.

5. SOMATIC HYBRIDIZATION: Somatic hybridization has been applied for improvement of cultivated plant species as genes can be transferred by protoplast fusion against bacterial, fungal and virus diseases or even nematodes and abiotic stresses such as drought, cold and soil salinity (Göntér *et al.* 2002). Somatic hybridization provides the breeders the possibility of accessing sexually incompatible genotypes between the crop species and distant relatives, merging genomes of sexually dysfunctional variety or breeding population and substituting one cytoplasm for another with a little effect on nuclear genome..

6.INTRAGENESIS AND CISGENESIS: Cisgenesis and intragenesis have been developed as novel tools aimed to modify crops. While cisgenesis involves genetic modification using a complete copy of natural genes with their regulatory elements that belong exclusively to sexually compatible plants, intragenesis refers to the transference of new combinations of genes and regulatory sequences belonging to that particular species.In cisgenesis, the gene of interest (i.e. cisgene), containing its native introns, promoters and terminator in sense orientation, is taken from the species itself or a sexually compatible relative for genetic transformation. So, the germplasm exploited in cisgenesis is identical to the gene pool available for traditional breeding. However, since foreign genes such as the selection marker genes are absent or eliminated from the primary intragenic/ cisgenic transformant or their offspring, the crops become far more acceptable by the general public. Cisgenic plants have no extra risks as compared to plants from conventional breeding or mutation breeding.

7. LINKAGE DRAG: One of biggest faced 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 (Friebe *et al*. 1996).

8. UTILIZATION OF GENE POOL DIVERSITY: For practical end products to be gained, some transfer prerequisites that encompass all three gene pool species are hybrid production, embryo rescue, plant regeneration, cytological diagnostics, breeding methodology and stress screening, culminating in the stability of the advanced derivatives contributed by homozygosity. Based upon these prerequisites and genetic transfer ease, primary gene pool diversity is most important for wheat improvement. The species of the diploid A and D genomes contribute novel genes and allow direct recombinational exchanges with their respective genome partners to facilitate both durum and bread wheat improvement over a relatively short-term time frame than what is provided by the secondary or tertiary gene pool species.

Detection of Alien Gene Transfer

In any alien gene transfer process where sexual hybridization is involved, it becomes very important to keep track of the validity of the wide hybrids and also the actual retention of the alien chromatin during generation advancement (Chaudhary *et al.* 2011). The extent and amount of gene transfer from wild relative into a crop species are determined by the breeding system of the plants (Hancock *et al.* 1996) as well as fulfilment of the necessary for hybrid formation and its survival in the environment.

Conclusions and Future Prospects

- Introgression in crop plants, mostly through conventional plant breeding and more recently aided by transgenesis and molecular marker technology, has proved to be one of the most powerful tools for plant breeding and has increased the yield levels of crop plants to the present level which is well supporting the ever-increasing population of the world.
- Future developments regarding the generation and commercialization of intragenic and cisgenic crops will depend on willingness to apply less stringent regulation to these crops worldwide. A less comprehensive regulation of intragenic/cisgenic crops, reducing the costs for approval, would be especially helpful to small-sized breeding and seed companies. This would provide these breeders with an additional tool for plant breeding and thus increase the number of intragenic/cisgenic crops developed.
- TILLING and EcoTILLING are cheap and rapid natural polymorphism detection and genotyping methods. They have advantages for determining the range of variation for genetic mapping based on linkage analysis.
- The plants have evolved in the nature along with the evolution of human civilization. While natural evolution has been governed by the environmental forces, the human interference has increased the process of evolution of crop species, particularly those which are of use to him either directly or indirectly.
- The transition of human race from a collector of seeds to a producer of grains led to more and more interaction between him and the plants which have been useful to him. This transition has travelled a very long way from unknowing attempts of domestication to crop breeding to genetic transformation culminating into present-day super domestication. Genetic diversity of crop plants, either natural or manmade, played significant role in this evolution.
- 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.

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