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Accelerated Backcrossing and Gene Pyramiding

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Breeders are often interested in transferring or introducing many genes from different sources into a desirable variety for genetic improvement. The process of combining traits is known as gene pyramiding, and the concept was proposed by Nelson (1978) to develop crop varieties with few to several different oligo genes for durable disease resistance. Gene pyramiding has also been called multitrait introgression, as often, genes governing two or more traits are introgressed into a single recurrent parent. Pyramiding of multiple QTLs/genes may be conducted through the multiple parent crossing, backcrossing. The end product of a gene pyramiding programis a genotype with all of the target genes. Since breeding is a very time-consuming process, breeders aim to combine as many desirable alleles as possible in a single breeding cycle (from crossing to the generation of near-homozygous breeding lines). When the number of genes to be assembled is known, the goal of gene pyramiding is to obtain near-homozygous breeding lines that are fully homozygous for the desirable alleles of the target genes using the minimum number of generations of selection and the lowest genotyping and phenotyping costs.

Important Traits to be Focused Upon

The first challenge in breeding for multiple traits is identifying and prioritizing those that are most important for the target environment and market. Although several traits of importance can be targeted simultaneously for the genetic improvement of cultivars, careful prioritization is essential because each expressed trait carries a biological and economic cost in the final phenotype. Breeders should therefore focus on identifying genotypes with desirable adaptation to biotic and abiotic stresses, superior grain quality, enhanced nutritional attributes, and appropriate phenology that matches the target environment.

Marker-Assisted Backcrossing/Accelerated Backcrossing-Basic Principles

Marker-assisted backcrossing (MABC) is a precise and effective method to introgress a single locus controlling a trait of interest while retaining the essential traits of the recurrent parent (RP). MABC is the process of using markers to select target loci, minimize the length of the donor segment containing a target locus, and/or accelerate the recovery of the RP genome during backcrossing.

The main objective of MABC is to integrate a targeted gene from agronomically substandard sources (the donor parent) into an elite breeding line (the RP). The anticipated product is an improved line comprising solely the targeted gene from the donor parent, with the remainder of the genome being that of the recipient parent. One of the early benefits envisioned with the use of molecular markers in plant breeding was their ability to accelerate the backcrossing process by reducing the number of backcrosses required to recover the RP phenotype. MABC is superior to conventional backcrossing in both precision and efficiency. Background selection can greatly accelerate the backcrossing program compared to conventional backcrossing alone.

MABC involves successive backcrossing to remove the genetic background of the donor while recovering the genetic properties of the RP as much as possible. MABC can also be used to develop near-isogenic lines (NILs) by minimizing carried-over donor segments flanking the target locus, thereby providing precise introgression of individual genes for detailed characterization of QTLs. The RP genome is recovered more slowly on the chromosome carrying the target locus than on other chromosomes because of the difficulty in breaking linkage with the target donor allele. However, by using large population sizes (400 or more plants) in backcross F_1 generations, it is possible to recover the RP genotype using only two or three backcrosses.

Foreground selection

Foreground selection, in which the breeder selects plants having the marker allele of the donor parent at the target locus. The objective is to maintain the target locus in a heterozygous state (one donor allele and one RP allele) until the final backcross is completed. Then, the selected plants are self-pollinated, and progeny plants are identified that are homozygous for the donor allele. Those markers that have already been developed and are tightly linked to the target gene or QTL should be used to select the target locus of the donor parent in early (BC) progenies for the selection of plants that have the target gene. This is referred to as “foreground selection,” although it is also referred to as “positive selection.” This may be particularly useful for traits that have laborious or time-consuming phenotypic screening procedures. It can also be used to select reproductive-stage traits at the seedling stage, allowing the best plants to be identified for backcrossing.

Background Selection

Except for the target locus, all genomic regions can be selected during background selection using recurrent parent (RP) marker alleles, while selection of the target locus is done based on phenotype. This selection is important to reduce unnecessary genes (linkage drag) introduced by the donor parent. By using molecular markers, it is easy to eliminate unwanted donor alleles present in the same genomic region as the target locus. The third level of MABC involves selecting backcross (BC) progeny with the greatest proportion of the RP genome using markers that are unlinked to the target locus; this process is referred to as background selection.

Molecular Approaches for Gene Pyramiding

Recurrent selection (RS) can fit population improvement by highly efficient pyramiding of different favorable alleles from diverse donors. Recently, multi-parent advanced generation inter-cross (MAGIC) populations have been introduced to the plant community. MAGIC populations are characterized by diversity in both alleles and phenotypes derived from multiple parents. In addition, relatively large recombination events within the population ensure high resolution of QTL mapping. Based on QTL mapping information and phenotypic performance, ideal individuals will be selected by pyramiding multiple favorable alleles in consecutive recurrent selection populations derived from inter-crossing among the top plants in each round of RS. MAGIC populations have been developed for the purpose of integrating QTL mapping with breeding in rice and wheat crops.

Discovery and introgression of a dominant male nuclear sterile (DMNS) gene into elite backgrounds make easy recombination of different alleles, producing half sterile individuals and another half fertile individuals in each round RS population. Human-aided out crossing between DMNS sterile plants and fertile plants greatly facilitates recombination of alleles at all loci among individuals in the progeny of the RS population. The strategy of molecular markers combined with RS, referred to as molecular recurrent selection (MRS), probably provides a very promising way to pyramid different favorable alleles (Figure 1). It should allow more effective identification of recombinants of favorable alleles at all target loci and more effective breakdown of genetic drag in the MRS population through DMNS-facilitated random mating.

Furthermore, genomic selection (GS) can be entirely integrated with RS. Bulk-harvested seeds from all fertile plants in the RS population after the third round of recombination could be used as a training population after genetic uniformity is achieved by rapid generation advancement (RGA) facilitation. Based on phenotypic data in the given environment and high-throughput SNP data of the training population, QTLs affecting the target traits will be identified and genetic prediction models will be set up for each target trait (Figure 1). In subsequent RS populations, the prediction model can be used to estimate genomic breeding values (GEBVs) of individuals for selection based on marker data, thus improving selection accuracy by avoiding phenotyping of the target traits.

Future Prospects

In breeding programs, it is necessary to develop crop varieties with multiple traits, including high yield and quality, as well as tolerance to abiotic and biotic stresses, in order to produce successful cultivars that meet consumer demands under challenging farming conditions. Conventional breeding methodologies for integrating multiple traits into a single genetic background have great potential; however, the process is often challenging. Breeders must prioritize target traits while considering the availability of resources, cost, and time required to breed for multiple traits.

Molecular breeding technologies, including marker-assisted selection, marker-assisted backcrossing, recurrent selection, and genomic selection, can greatly supplement conventional breeding approaches and facilitate the integration of multiple traits or genes. Accelerated backcrossing and genomic selection, which represent the future of molecular breeding tools, have great potential to increase the efficiency of selection and improve multiple traits simultaneously.

Molecular breeding tools are still in their infancy for many crops. Therefore, there is a continuous need to develop effective genomic resources for both major and minor crops, including high-throughput marker identification and development, high-density linkage maps, and gene identification methodologies.

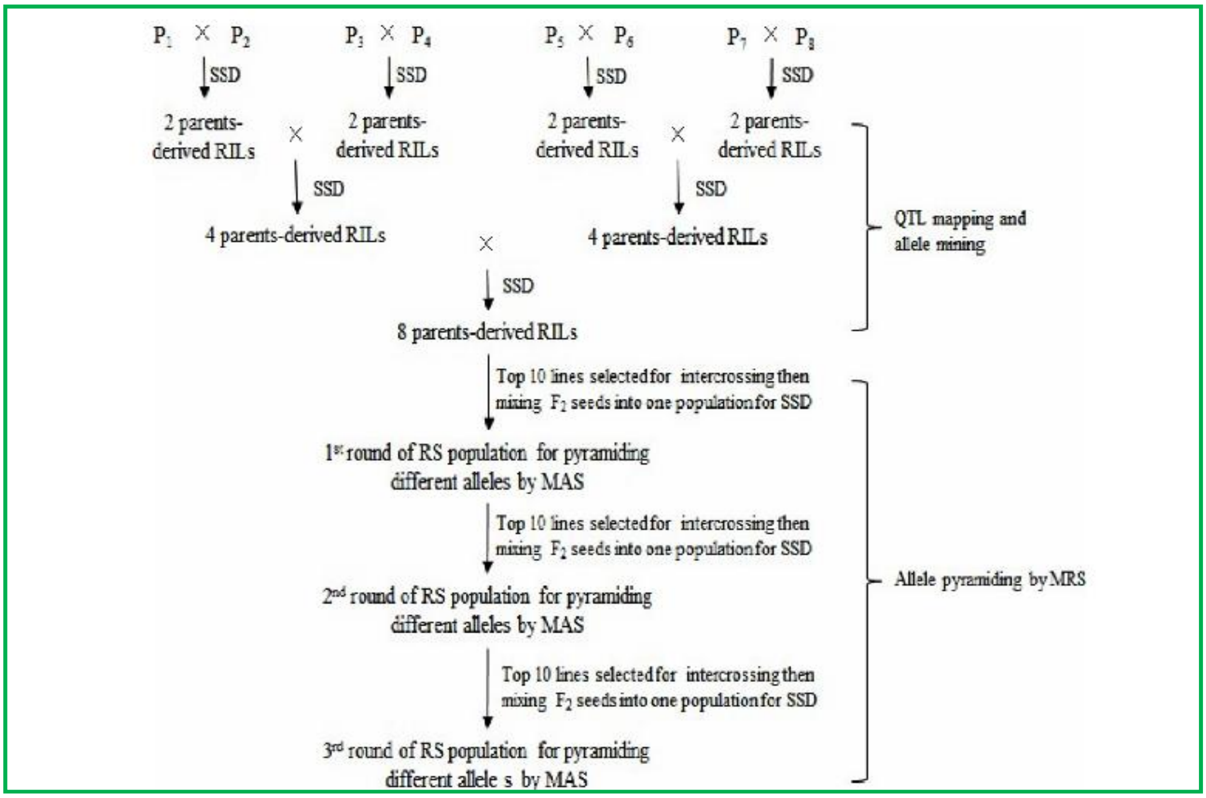

Figure 1 . Development of MAGIC population and its derived recurrent selection (RS) population for allele pyramiding by MAS.SSD: single seed descent, MRS: molecular recurrent selection

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