

MODERN TECHNOLOGIES TO IMPROVE CROP PLANTS

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Abstract In plant science selection plays a key breeding approach for improving economic features in many plants' kind. Selection techniques vary based on the mechanism of reproduction. Self-pollinated crops employ pure line and mass selection methods, whereas cross-pollinated crops use recurrent selection. Vegetative propagated plants use clonal selection. Regardless of Phenotypic recurrent selection is a successful approach for enhancing polygenic features. While increasing the frequency of desirable genes for economic characters can help maintain high genetic variability in heterozygous populations, the efficiency and effectiveness of this approach may not be satisfactory in most cases. This is due to phenotypic selection being influenced by environmental factors and genotypic selection taking at least 2-3 crop seasons per cycle. Molecular marker technology allows for faster and more efficient selection, as it is unaffected by environmental factors. Marker Assisted Recurrent Selection is a key strategy in molecular breeding because it allows for multiple QTLs to control complex traits, unlike Marker Assisted Selection and Marker Assisted Backcrossing.

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Introduction

Plant breeding is the method of developing superior varieties of plants with desirable characteristics from existing ones. Plant selection, introduction, and hybridization are among the breeding methods used to create superior varieties with economically valuable features. Among them, selection is an essential breeding approach used to increase a variety of economic features in plants. Selection is split into two major categories based on its source: natural selection and artificial selection. In natural selection, Superior plants are picked by natural forces such as rainfall, temperature, biotic and abiotic agents, with no human intervention (Abbas et al., 2024; Arshad et al., 2024; Junaid and Gokce, 2024). Here, all of the plants in the population are permitted to reproduce, preserving the population's genetic variety. In the instance of artificial selection, the superior plants in the population are chosen by human intervention. The selected plants are subsequently exposed to a variety of breeding processes, depending on how they reproduce. In autogenous crops, pure line and mass selection methods are adopted to develop

homozygous pure line varieties. Phenotypic recurrent selection is the process of reselection generation after generation, followed by interbreeding among chosen plants to create the population for the following cycle of selection. As a result, it entails selection cycles, recombination, and assessment aimed at raising the frequency of the favourable allele in the population. As a result, it is regarded as a useful technique for improvement in polygenic features by raising the frequency of desirable or advantageous genes for a variety of economic characteristics. It also assists in maintaining high genetic variety by recurrent inter-mating among the chosen plants of the heterozygous population. In vegetative propagated plants, clonal selection is used to isolate the best clones from progenies. Today, most cultivated plants are the result of artificial selection (Fatima et al., 2023; Haider et al., 2023; Rasheed et al., 2024).

However, because environmental variables influence genotype phenotypic selection and genotypic selection might take longer—at least two to three crop seasons for a single cycle of selection—the efficacy

and efficiency of selection are often insufficient. Because the environment does not affect the selection process, the molecular marker method might potentially speed up or enhance it. Among the various molecular marker technology, MARS is a potential one.

In this review, we will discuss Marker Assisted Selection, specifically Marker Assisted Recurrent Selection (MARS), and its relationship with other molecular breeding strategies.

Molecular Breeding Approaches

The phrase molecular breeding refers to several modern plant breeding procedures, including

- Marker-assisted selection (MAS),
- Genomic selection (GS).
- Marker-assisted backcrossing breeding (MABB),
- Marker-assisted recurrent selection (MARS),

A selection approach known as "Marker Assisted Selection" (MAS) uses linked markers to indirectly choose a genotype that harbours desired genes. Based on the marker system, uses the linked markers of DNA banding pattern on a gel or autoradiogram to indirectly evaluate the presence or absence of a desired plant characteristic ([Rani et al., 2014](#)). The idea is that the banding pattern revealing the paternal lineage of the bands in sergents at a given marker locus indicates the lack or presence of a certain chromosomal segment containing the targeted allele ([Rani et al., 2014](#)). MAS is becoming important in modern times because it helps and enhances the effectiveness of plant breeding by keeping an eye on whether advantageous genes are present in populations. This is accomplished by carefully moving genomic regions of interest and speeding up the recurrent parent genome's recovery. This has been more commonly used for simply inherited features (monogenic) than for polygenic traits (controlled by multiple genes, each with tiny impacts).

MABB is the most basic type of MAS, to introduce a significant gene from a disease-resistant plant into an exceptional breeding line. When numerous genes or QTLs are responsible for the expression of complex characteristics, emerging techniques like GS and MARS can be more efficient since they enhance accuracy, and reduce the cost of phenotyping, and time ([Bernardo, 2010](#)). A kind of MAS known as "genomic selection" uses genetic markers spread throughout the whole genome to guarantee As a result, it helps improve quantitative characteristics of plant populations.

Marker Assisted Recurrent Selection (MARS)

Marker-aided Recurrent Selection (MARS) is type of a recurrent selection technique that employs molecular markers to locate and select multiple genomic regions that are involved in the development of complex characteristics for assembling the most effective genotype within a single population or across related populations ([Ribaut et al., 2010](#)). MARS is a system, that makes it possible for selection

on base of genotypic and intercrossing between the chosen plants in the same crop season for a single cycle ([Ribaut et al., 2010](#)). As a result, MARS may improve effectiveness in recurrent selection and speed up the process ([Gazal et al., 2015](#); [Ragot et al., 2000](#)). MARS has been proposed for "forward breeding" of native genes and pyramiding numerous QTLs to address complex variables ([Jiang et al., 2007](#)). MARS performs QTL mapping for each breeding population. This allows for the incorporation of many QTL with minor genetic contributions that impact the expression of complex characteristics. Once the critical QTLs of importance have been identified in the MARS breeding population, the selected individuals are subjected to controlled pollination to produce lines with the best compliment of QTLs from both parents ([Crosbie et al., 2006](#); [Eathington, 2005](#)).

Methodology of MARS

MARS consists of estimating the effects of marker using genotyping and phenotyping of F2 or F3 generated F4 or F5 progenies, followed by two to three cycles based on the presence of marker alleles for modest impact QTLs. The initial phase in MARS involves fingerprinting the children of a particular breeding population resulting from a biparental cross using specified molecular markers ([Eathington et al., 2007](#)). This implies that QTLs are found in 200-300 or more progenies from the F3 population using particular molecular markers, allowing a genotypic value to be calculated for progeny. In the next stage, around 250-280 progenies from the F-3 population (F3:4 or F3:5) are examined at many locations to collect phenotyping data. Based on genotyping and phenotyping data, a small number of plants are chosen and allowed to pollinate or interminate for two or three cycles. Using continuous nursery programs and pre-flowering information, many cycles of molecular MAS and controlled pollination may be completed in one year. ([Eathington et al., 2007](#); [Gazal et al., 2015](#)) Following that, the recombined lines (F3 or F3:4) are obtained through continuous selfing for two or three years from the produced material ([Eathington et al., 2007](#); [Gazal et al., 2015](#)).

Multiallelic MARS

To maximize genetic gain, MARS has demonstrated its ability to integrate advantageous alleles for characteristics of interest that are distributed across several parents into a single inbred line ([Ooijen and Jansen, 1994](#)). This may be accomplished by breaking up the multi-allelic scenario into several bi-allelic circumstances, and then combining the results of these bi-allelic MARS to create inbred lines ([Peleman and Van der Voort, 2003](#)). If there are few target loci, using partial backcrossing or MABC to successively pyramid favourable alleles from several sources into a single genotype may be a better option than using MARS. However, breeding with several sources of advantageous alleles simultaneously, as opposed to carrying out sequential bi-allelic line development programs, might save a great deal of time and hence

increase the pace of genetic gain ([Ribaut and Ragot, 2007](#)).

MARS and Phenotypic Recurrent Selection

Phenotypic recurrent selection selects plant genotypes depending on their phenotype traits. Chosen individuals are self-tested while also being tested by a tester. Selfed seeds from chosen plants are gathered and kept independently for use in the third year. The test cross progenies are examined in replicated yield trials, and the best test cross progeny are chosen. The selfed seeds from the plant that produces better test cross offspring are planted in crossing blocks to provide for the most potential intercrosses. Then, an equal number of seeds from each intercross are collected to produce the following generation of the cycle. This entire process consists of one cycle, which can be repeated twice or three times to raise beneficial allele frequency in that population. Regardless of having greater potential for improving several agronomical characteristics, selection efficiency is not always suitable in some cases because environmental factors heavily influence phenotypic selection and genotypic selection takes a long time, with at least two to three crop seasons required for each cycle of selection ([Bernardo and Charcosset, 2006](#)).

In contrast, in (MARS), plant genotypes are chosen based on particular markers connected to genes, and intercrosses between selected individuals are also made within the same crop season for one cycle of selection. As a result, the selection process can be more efficient, and the integration of beneficial genes can move more quickly ([Eathington et al., 2007](#); [Gazal et al., 2015](#)). Several scholars have already demonstrated the superiority of MARS over the phenotypic recurrent selection strategy. Several computer simulations of MARS and phenotypic recurrent selection have shown that MARS is typically more effective than phenotypic selection in consolidating favourable genes from several parents into a single offspring. However, the benefit of MARS over phenotypic selection was greatest when the population under selection was extremely varied, as materials geographically apart from one another ([van Berloo and Stam, 1998](#)). Further simulation experiments revealed that the comparative advantage of MARS over phenotypic selection diminishes rapidly as the proportion of total genotypic variation described by QTLs included in the selection index decreases ([Moreau et al., 2004](#); [Openshaw and Frascaroli, 1997](#)) observed that the genetic gain acquired via MARS was greater than the phenotypic recurrent selection ([Charmet et al., 1999](#); [van Berloo and Stam, 1998](#)). Similarly, the genetic gain produced using MARS in maize was nearly double that of phenotypic selection (PS) in certain populations. Hence, MARS is unquestionably a better alternative for the accumulation of favourable genes from parents in a single genotype ([Crosbie et al., 2006](#); [Eathington, 2005](#)).

MARS and MABC

One or a few QTLs with a significant genetic impact from an agronomically inferior genotype but with another desired characteristic (a non-recurrent parent) can be successfully transferred into an elite cultivar using the MABC method. However, it is challenging to combine all advantageous alleles in one genotype by MABC for the majority of complex variables. On the other hand, the MARS method, which uses multi-trait selection indices and identification and crossover among chosen individuals for several generations based on their molecular marker genotypes, allows one to approach or even reach a perfect genotype. ([van Berloo and Stam, 1998](#)) ([Peleman and Van der Voort, 2003](#)). The benefit of the MARS system is that it targets large QTLs by capturing several genomic areas. It tries to use markers that are strongly linked with desired traits. Consequently, the resultant genetic gain from the MARS program is higher than that of the MABC program ([Bernardo, 2008](#); [Bernardo and Charcosset, 2006](#)).

MARS and Genomic selection (GS)

Genomic selection is a marker-based approach that builds a model to forecast the genetic value of progenies for selection by using all the molecular marker data that is currently accessible, encompassing the entire genome at once ([Lorenz, 2013](#); [Meuwissen et al., 2001](#)). By treating every marker as a potential QTL and ensuring that every gene or QTL in the whole genome is in linkage disequilibrium with at least one marker, this approach lowers the possibility of overlooking small-effect QTLs ([Guo et al., 2012](#)). According to ([Bernardo and Yu, 2007](#)) using all of the molecular markers that span the entire genome, as opposed to just the subsets of markers that are strongly linked to QTLs, improved breeding value prediction and accuracy, according to computer simulation research. In a similar vein, 284 markers were used for genotype 233 recombinant inbred lines (which were produced by crossing B73 and Mo17 parents in maize) ([Massman et al., 2013](#)). The results showed that GS outperformed MARS. While the GS method is more effective than MARS in locating small impact QTLs throughout the whole genome, this is only the case when all of the plant species' genome molecular markers are utilized ([Nakaya and Isobe, 2012](#)). Consequently, it has not gained traction in plant breeding, which may be attributed to a lack of useful knowledge on whole genome molecular markers. In contrast, in MARS, a subset of molecular markers may be used to find genotypes that show promise and have desired QTLs or small-impact genes in the population. These genotypes can then be integrated from multi-parent genotypes into a single genotype by intermingling with one another. Therefore, MARS is superior to GS when information is available on a restricted group of markers as opposed to molecular manufacturers encompassing the entire genome.

Practical utility of MARS

MARS was first created for large-scale commercial breeding programs of maize to identify and recombine progenies in breeding populations that have the best QTLs from either parent (Johnson, 2004; Ragot et al., 2000). The rate of genetic gain in commercial maize was subsequently doubled as a result of this technique. Private maize breeding initiatives have successfully used MARS in several large-scale and practical research (Crosbie et al., 2006). All generations of MARS, including the two population parents, had their genetic values estimated for both the index and individual characteristics. The results showed that, when choosing people based on an index, the genetic values of individual traits increased in the intended direction. Similar to this, Johnson, (2004) carried out an experiment (the maize MARS project) in which he showed the effectiveness of MARS for quantitative features using large-scale molecular markers in bi-parental populations (Ragot et al., 2000). To maximize the effectiveness of long-term selection by raising the frequency of advantageous alleles, these markers are first utilized for genes or QTL identification and then for MARS on yield (i.e., quick cycles of selection and recombination based on related specialized markers). In (Eathington et al., 2007) study, MARS lines exhibited superior performance compared to traditional chosen lines in Monsanto's Brazilian maize breeding program. According to (Bernardo, 2008), MARS raised the frequency of the favorable marker allele at 18 of the 31 markers used in the first sweet corn F2 population and at 11 of the 35 markers used in the second sweet corn F2 population, from 0.50 to 0.80. The International Maize and Wheat Improvement Centre (CIMMYT) in Hyderabad is home to one of the research centers. It is possible that this centre was the first in the public sector to successfully use the MARS technique to develop early-generation yellow drought-tolerant maize inbred lines for the AMDROUT (Asia Maize Drought Tolerance) project.

This method has been used in other crops, such as sunflower and soybean, in addition to the maize breeding program, to combine beneficial alleles at many target loci distributed across several parents into a single elite line. In comparison to traditionally selected lines, (Eathington et al., 2007) showed improvements in grain moisture at harvest, grain yield, and percent oil in European sunflower breeding populations. In as many as 40 soybean breeding populations, the MARS lines demonstrated a 37.6 kg per ht yield advantage with a slight delay in relative maturity. According to (Yi ChengXin et al., 2004), improved populations of upland cotton had a mean degree of resistance to *Helicoverpa armigera* that was considerably greater than that of the previous population following MARS. Thus, MARS can help improve quantitative characteristics such as grain moisture and yield, among others. Selection enhances the commercial qualities of several plant species

variations. Recurrent selection is one of the many selection techniques used in plant breeding that is crucial to population development because it increases the frequency of advantageous alleles in the population. However, in certain cases, the efficiency of this process is not sufficient since genotypic selection takes longer—two to three crop seasons, at least—for a single cycle of selection, while phenotypic selection is heavily dependent upon a variety of environmental factors. To address these issues, MARS is superior to other molecular plant breeding techniques like MAS, MABB, and GS, especially when several genes or QTLs are controlling the expression of several complex variables like yield, biotic, and abiotic traits that have little genetic impacts. This approach was first created to select and recombine progenies with optimal QTLs from either parent in breeding populations for large-scale commercial maize breeding programs. However, it is currently being used to enhance economic features in a variety of crops, including cotton, sunflower, soybean, and so on. Therefore, MARS can help improve a variety of quantitative characteristics, such as grain yield and moisture, disease resistance, and drought resistance, in a variety of crops. MARS can integrate numerous favourable genes along with the selection of a complicated feature.

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Author Contribution statement

All authors contributed equally.

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All data generated or analyzed during the study are included in the manuscript.

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Conflict of Interest

Regarding conflicts of interest, the authors state that their review was carried out independently without any affiliations or financial ties that could raise concerns about biases.



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