Crop improvement through Marker Assisted Selection

Patel Savankumar N., Assistant Professor (GPB), School of Agriculture, P P Savani University, Surat - 394125.,

 Sakriya Shveta G., Assistant Professor (Horti.), School of Agriculture, P P Savani University, Surat - 394125.,

Amol Langhi, Junior Research Assistant, Agricultural Research Station, Savalivihir Farm, MPKV, Ahmednagar- 423109.

Ketan S. Mungra, Assistant Research Scientist, Main Rice Research Center, Navsari Agriculture University, Navsari-396450.

**ABSTRACT**

Marker-assisted selection (MAS) is a powerful and successful strategy used in crop development to accelerate breeding and improve trait selection. It is the process of selecting agriculturally important traits for crop development by using morphological, biochemical, or DNA markers as indirect selection criteria. Traditional breeding processes are usually labour- and time-intensive, with little guarantee of obtaining the desired attributes. However, the development of molecular markers and advances in genomics have fundamentally altered the area of plant breeding. By discovering and utilising markers that are highly linked with the trait of interest, breeders can drastically reduce the time of breeding cycles. In this study, we present an overview of the benefits of MAS and its most widely used applications.

**Keywords**: Marker assisted selection, Crop improvement, Trait

1. **INTRODUCTION**

Marker-assisted selection (MAS) is a sophisticated and powerful strategy used in crop improvement programmes to speed breeding and enhance the efficiency of developing improved plant varieties. Traditional breeding processes can be time-consuming and labor-intensive, requiring multiple generations to achieve desired crop characteristics. One sector of biotechnology with significant potential for plant breeding is DNA marker technology, which emerged from studies in molecular genetics and genomics. DNA markers can be used to detect allelic variation in the genes underlying these traits because of genetic linkage. The use of DNA markers to aid in plant breeding could improve efficiency and precision significantly. Marker-assisted selection (MAS) is a component of the burgeoning discipline of "molecular breeding." It involves the use of DNA markers in plant breeding. Collard and colleagues (2008). The primary goal of crop improvement with MAS is to choose plants with specific desirable molecular characteristics. These characteristics could be related to productivity, disease resistance, stress tolerance, nutritional content, or any other economic or agronomic value. Traditional breeding takes time and is heavily reliant on environmental factors. A new variety takes between eight and twelve years to breed, and even then, the emergence of an improved variety cannot be guaranteed. As a result, breeders are keenly interested in emerging technology that could improve the process's efficiency. Molecular marker technology offers such promise by applying a variety of novel ways to improve selection strategies. (Akhtar and colleagues, 2010). A basic issue that must be addressed is how to improve the efficacy of marker-assisted selection for quantitative traits by characterising target genes effectively. Fortunately, over the last decade, field design and statistical approaches for QTL mapping have evolved significantly. Using the most recent mathematical approaches, such as composite interval mapping, field data from various settings can be combined into a joint analysis to evaluate the QXE and so identify 'stable' QTLs across environments. Furthermore, when combined with a comprehensive linkage map, composite interval mapping enables for more precise identification of QTLs in the genome as well as improved identification of coupled QTLs (linked QTLs in which the favourable allele is inherited from the same parental line [3].

**RESULTS AND DISCUSSION**

Marker-assisted selection has been applied successfully in numerous crops, including rice, maize, wheat, and soybeans. Its combination with traditional breeding methods has resulted in the development of enhanced crop varieties that address the challenges of modern agriculture, such as climate change, pest and disease risks, and rising food security demands.

The late-maturing high-yielding rice variety reeta from the donor parent, Swarna-Sub1, was introgressed with the quantitative trait loci (QTLs) Sub1 for submergence and Pup1 for low phosphorus stress tolerance as well as the narrow-grained trait GW5. The generated lines shared all morphoquality characteristics with the receiving parent, were better yielding, possessed submergence, and had low phosphorus stress-tolerance. Reeta-Panidhan (CR Dhan 413), a prospective pyramided line, has been released for the flood-prone areas of Odisha state using marker assisted breeding [4].

The use of MAS large embryos and golden-like characteristics in coloured rice. It demonstrates that the combination of molecular markers and traditional breeding methods may be utilised efficiently for biofortification. PFR32 and RFR13 have the potential to become profitable products that can adapt to the existing agricultural community [5].

Tomato bacterial, fungal, and viral infections are to blame for widespread output losses, especially in humid growth conditions. Marker-assisted selection was used to look for unusual recombination events that linked these resistance loci into a connected cassette that can be inherited collectively in subsequent crosses. Using a pedigree breeding strategy and marker-assisted selection, researchers discovered a new link between Xv3/Rx4 and Ty-2. Because they are closely associated to the appropriate resistance loci, the trait markers on chromosome 11 are effective for selecting plants that are resistant to the target illnesses, according to the findings of this study [2].

The use of markers in selection (MAS) allows breeding to be accomplished faster and with fewer resources. Novel QTLs and SSR markers can be used to improve significant fibre quality parameters such as fibre strength, fibre length, fibre uniformity, and micronaire. The new cultivars "Ravnaq-1/Ravnaq-2," which were registered with the State Variety Testing Commission of Uzbekistan in 2014-2017, are the first generation of MAS-derived cotton cultivars in Uzbekistan. These findings show the utility of MAS in cotton breeding by demonstrating how the LD-block of chromosome 7 and its mapped molecular marker(s) and donor genotypes effectively assisted in the accurate and rapid transfer of higher fibre quality QTLs to commercially cultivated Upland cotton cultivars [6].

To identify the genetic loci for cold tolerance and to establish a molecular marker-assisted selection (MAS) method, 23 effective cross combinations were obtained through a half diallel cross among 16 parents, including eight frost-tolerant advanced breeding lines and eight cultivars. Bulked segregant analysis (BSA) was used to map three putative frost tolerance areas on chromosomes II, V, and IX. Furthermore, six SNP markers related with frost resistance were created and verified from candidate locations. Above all, a MAS system was built for cold tolerance screening of early breeding offspring. The study emphasises the practical benefits of employing diallel populations to widen and improve frost-tolerant genetic resources in potato via marker assisted selections [7].

Marker assisted selection can be used effectively in wheat resistant breeding to control stripe rust. Gene pyramiding is thought to be an effective method for increasing long-term disease resistance in wheat. In this experiment, a panel of Yr gene pyramiding lines (containing of 3-8 Yr genes) were created using marker aid selection using cv. Chuanyu12 as the background parent and evaluated under currently epidemic Pst races. The quantity of pyramided Yr genes was found to be significantly linked with stripe rust resistance (p0.001). The current Pst races benefit from Yr15, Yr62, and Yr65. More than four effective or partially functional Yr genes stacked together can provide enough resistance to stripe rust. The quantity of pyramided Yr genes was found to be significantly linked with stripe rust resistance (p0.001). The current Pst races benefit from Yr15, Yr62, and Yr65. Furthermore, the impacts of Yr gene pyramids on currently prevalent Pst races and agronomic qualities were assessed, and Yr-gene pyramiding lines with desirable agronomic features were developed for long-term Pst control in wheat breeding [8].

The biotrophic fungus Blumeria graminis (DC.) Speer f. sp. tritici emend. E.J. Marchal is the cause of powdery mildew, one of the most damaging diseases that endangers the security of common wheat production. At the seedling stage, this disease primarily manifests as white colonies on the leaf; later on in the growth stage, it also appears on spikes. B. graminis f. sp. tritici infection lowers chlorophyll concentration, interferes with photosynthesis, and usually reduces wheat production by 10–15%, and in extreme cases, up to 62%. There will be an impact on flour quality as well. The production of disease-resistant cultivars and parental selection benefit from the characterization of powdery mildew resistance genes. A Chinese wheat breeding line known as KN0816 shows better agronomic performance and resistance to powdery mildew resistance at all growth stages.

Genetic analysis of KN0816 populations crossed with susceptible parents indicated that seedling resistance to different isolates of B. graminis f. sp. tritici was conferred by a single dominant gene, provisionally designated PmKN0816. Using polymorphic markers linked to the catalogued genes Pm6, Pm52, and Pm64, PmKN0816 was mapped to the Pm6 interval on chromosomal arm 2BL. It was surrounded by the markers CISSR02g-6 and CIT02g-2, both of which had a genetic distance of 0.7 cM. An examination of closely related molecular markers revealed that the marker alleles of PmKN0816 were distinct from those of other powdery mildew resistance genes on 2BL, such as Pm6, Pm33, Pm51, Pm64, and PmQ. Based on its genetic and physical locations as well as its reaction pattern to different B. graminis f. sp. tritici isolates, PmKN0816 is most likely a new powdery mildew resistance gene with effective resistance to all 14 studied isolates. Due to KN0816's combined resistance and better agronomic performance, PmKN0816 is expected to play a significant role in wheat breeding. In order to use marker- aided selection to transfer PmKN0816 to different genetic backgrounds, closely associated markers were evaluated. Four of these markers (CIT02g-2, CISSR02g-6, CIT02g-10, and CIT02g-17) were found to be suitable for MAS in various genetic backgrounds [9].

Agriculture using sugar beetroot (Beta vulgaris L.) is threatened by Rhizoctonia solani, a disease that causes rot in the roots and crown of the plant. The development of resistant cultivars with the use of marker-assisted selection is given top priority in breeding efforts. Restrictions site-associated DNA sequencing of two geographically distinct sets of plant materials with varying degrees of resistance/susceptibility resulted in the identification of a single-nucleotide polymorphism marker linked to Rhizoctonia resistance, allowing for a more diverse selection of superior genotypes. The raw SNPs obtained by RAD sequencing (15,988 and 22,439 SNPs) explained 13.40% and 25.45% of the phenotypic variability in the two sets of material from different sources of origin, respectively. The SAMtools variant calling approach was applied. On the basis of their contribution to the phenotype, mutually occurring significant SNPs were filtered using principal component analysis (PCA) biplots after an independent association analysis on both datasets was completed. High-resolution melting, Sanger sequencing, and rhAmp SNP genotyping were combined in a systematic molecular validation process to produce a ready-to-use marker for the breeding community. These SNPs were distributed across the genome. We report that 10% of the variation in phenotypic disease is explained by RsBv1, which is found on Chromosome 6 (9,000,093 bp) and is significantly associated with Rhizoctonia resistance (p < 0.01). In order to breed sugar beets resistant to Rhizoctonia, the corresponding SNP assay is now prepared for marker-assisted selection [12].

Marker-assisted selection is a common technique in tomato resistance breeding. Under controlled experimental circumstances, 964 tomato lines were screened extensively to test the resistance of major molecular markers often used in MAS. Initially, 36 molecular markers were examined against 26 resistance genes (R genes) and 14 major illnesses. They analysed the data using traditional molecular biology and bioinformatics methods, with polymorphism, accuracy, and clarity of amplicons acting as marker selection criteria. Following an initial review, 20 of these markers were recognised as efficient markers, with 8 of them being gene-based markers and referred to be ideal markers. 18 R genes that control 12 diseases were identified as efficient markers in the PCR data. Grouping breeding lines based on the amount of R genes carried, on the other hand, revealed that 62% of the lines were empty of R genes, while 38% had many types of R genes. This allows for a better understanding of novel sources of resistance in breeding lines. Finally, these efficient markers and their restricted PCR conditions can be proposed as the basis for a diagnostic kit for MAS applications against 12 major tomato diseases, and the identified resistant breeding lines can be conserved and propagated as different sources of resistance for the development of new resistant varieties. As a result, high efficiency combining of the essential R genes and pyramiding into commercial tomato varieties are advocated as a pragmatic strategy in disease-prone locations [13].

Sorghum is an important staple food crop in Sub-Saharan Africa's drought-prone areas, where rainfall is unreliable and unevenly distributed. When compared to other cereal crops, sorghum is a drought-tolerant crop with a fair yield, albeit abiotic stress reduces productivity. Stay-green sorghum cultivars maintain green functioning leaves throughout post-anthesis drought stress, making it a valuable crop for food and nutritional security. Nonetheless, because of climate change driven by human activities, it is difficult to maintain tolerance over time. Drought is addressed in sorghum by a variety of techniques, including breeding drought-tolerant sorghum with traditional and molecular technologies. Traditional techniques have the disadvantage of relying on phenotyping staygreen, which is challenging in sorghum because to the presence of multiple genes and environmental variables. Marker assisted selection, which uses DNA molecular markers to identify QTL associated to stay-green improvement in sorghum, has been shown to be helpful in enhancing stay-green improvement. It requires comparing phenotypic field data with QTL mapping associated with the stay-green trait for introgression into senescent sorghum varieties. In Tanzania, researchers used marker-assisted backcrossing to improve the stay-green (STG) qualities of farmer-preferred sorghum cultivars. Using previously published KASP markers related with STG 3A and STG 3B QTL, 752 individuals from five BC2F1 populations and their parents were genotyped. In the BC2F1 populations, the S35\*Pato background had the most individuals with heterozygous alleles (37) while only seven individuals descended from the B35\*Wahi parents' background had heterozygous alleles. Beneficial alleles were detected in 18 of the 30 single nucleotide polymorphism (SNP) markers in BC2F1 populations. In the BC2F1 generation, the B35\*NACO Mtama 1 background population had the highest grain yield (0.127 kg/panicle). Genotypic analysis revealed the presence of beneficial alleles in homozygous conditions at markers loci related with STG 3A and STG 3B QTLs in BC2F3 populations, showing successful introgression of STG QTLs from donor parents to recurrent parents. Across all water irrigation regimes, the genotype NA316C exhibited the highest mean grain weight (0.068 kg/panicle). As a result of our findings, we can use marker-assisted backcrossing to improve drought tolerance in African sorghum varieties [14].

Maize is an excellent source of nutrients and is a staple meal in many countries of the world, including India. Creating a maize genotype with increased levels of lysine and tryptophan, as well as -carotene, can help reduce protein-energy malnutrition (PEM) and vitamin A deficiency (VAD). To increase lysine and tryptophan content by transferring the opaque-2 (o2) gene from donor HKI163 to -carotene-rich inbred lines, UMI1200+ and UMI1230+. F1, BC1F1, BC2F1, BC2F2, and BC2F3 plants were created with an o2 line HKI163 and two -carotene-rich inbred lines, UMI1200+ and UMI1230+ as parents. The target genes were chosen via foreground selection with the related marker umc1066 for the o2 gene and the marker crtRB1 3′TE for the crtRB1 gene. The background selection used a total of 236 simple sequence repeat (SSR) markers spread uniformly across the maize genome. Individual plants homozygous at the crtRB1 locus and heterozygous at the o2 locus were selected and backcrossed to produce BC2F1 plants to fix the crtRB1 allele in the BC1F1 stage. In addition, the selected heterozygous BC2F1 plants from both crossings were selfed to produce BC2F2 plants, which were then selected for the target gene and selfed to produce BC2F3 lines. Five improved lines with homozygous marker alleles for the crtRB1 and o2 genes were found from each cross, with RPG recovery ranging from 86.75 to 91.21% in UMI1200+HKI163 and 80.00 to 90.08% in UMI1230+HKI163. The enhanced lines performed well agronomically and exhibited high levels of lysine (0.294-0.332%), tryptophan (0.073-0.081%), and -carotene (6.12-7.38 g/g). These improved lines have the potential to be utilised as genetic resources for maize improvement [15].

**CONCLUSION**

## It is important that plant breeding continue to make incredible strides in crop enhancement. Traditional breeding practises appear to be being used in modern breeding operations to advance them. Although there hasn't been much of an impact on variety development thus far, MAS has the ability to significantly help plant breeders achieve this goal. To realise the potential of MAS, greater breeding programme integration, a comprehensive understanding of current restrictions, and the creation of realistic solutions are required. Using the benefits of MAS above traditional breeding could have a major impact on the battle against biotic and abiotic stressors, as well as yield and quality improvements. In the foreseeable future, the high cost of MAS will continue to be a substantial obstacle to its deployment for some crop species and plant breeders in developing countries. Customised MAS approaches may be required to accommodate specific crops, traits, and cost constraints. With new marker technology, the price of MAS could be drastically decreased. If the usefulness of the new methodologies is proved and the necessary equipment is readily available, MAS should be more widely used for crop improvement

## .REFERENCES

1. B.C. Collard and D.J. Mackill “Marker-assisted selection: an approach for precision plant breeding in the twenty-first century.” Philosophical Transactions of the Royal Society B: Biological Sciences. 2008, 363(1491), 557-572.
2. C.J. Orchard, J. Kressin, S. Chompookam, J. Chuapong, N. Onmanee, K. Van Leeuwen and D.M. Francis. “Marker-assisted Selection to Combine Alleles for Four Disease Resistance Genes of Tomato Collocated on Chromosome 11.” HortScience, 2023, 58(5), 495-501.
3. S. Akhtar, M.A. Bhat,, S.A. Wani, K.A. Bhat, S. Chalkoo, M.R. Mir, and S.A. Wani “Marker assisted selection in rice. Journal of Phytology” 2010, 2(10).
4. S.R. Barik, A. Moharana, E. Pandit, A. Behera, A. Mishra, S.P. Mohanty and S.K. Pradhan. “Transfer of Stress Resilient QTLs and Panicle Traits into the Rice Variety, Reeta through Classical and Marker-Assisted Breeding Approaches.” International Journal of Molecular Sciences. 2023, 24(13), 10708.
5. Y.P. Wu, Y.C. Chang, H.I. Kuo, B.N. Lin, S.M. Wang, and Y.C. Tseng. “The development of two high-yield and high-quality functional rice cultivars using marker-assisted selection and conventional breeding methods.” International Journal of Molecular Sciences. 2022, 23(9), 4678.
6. M.M. Darmanov, A.K. Makamov, M.S. Ayubov, N.N. Khusenov, Z.T. Buriev, S.E. Shermatov, ... and I.Y. Abdurakhmonov. “Development of superior fibre quality upland cotton cultivar series ‘Ravnaq’using marker-assisted selection.” Frontiers in Plant Science. 2022, 13, 906472.
7. W. Tu, J. Li, J. Dong, J. Wu, H. Wang, Y. Zuo, ... & B Song. “Molecular Marker-Assisted Selection for Frost Tolerance in a Diallel Population of Potato.” Cells. 2023, 12(9), 1226.
8. R. Liu, J. Lu, M. Zhou, S. Zheng, Z. Liu, C. Zhang, ... & L. Zhang, Developing stripe rust resistant wheat (Triticum aestivum L.) lines with gene pyramiding strategy and marker-assisted selection. Genetic Resources and Crop Evolution. 2020, 67, 381-391.
9. W. Wang, H. He, H. Gao, H. Xu, W. Song, X. Zhang, ... & P. Ma. Characterization of the powdery mildew resistance gene in wheat breeding line KN0816 and its evaluation in marker-assisted selection. Plant Disease. 2021, 105(12), 4042-4050.
10. H.J. Li, T.D. Murray, R.A. McIntosh, and Y Zhou. Breeding new cultivars for sustainable wheat production. Crop J. 2019, 7, 715-717.
11. R.P. Singh, P.K. Singh, J. Rutkoski, D.P. Hodson, X.Y. He, L.N. Jørgensen, M.S. Hovmøller, and J. Huerta-Espino. Disease impact on wheat yield potential and prospects of genetic control. Annu. Rev. Phytopa- thol. 2016, 54:303-322.
12. S. Ravi, M. Hassani, B. Heidari, S. Deb, E. Orsini, J. Li, ... & P. Stevanato. Development of an SNP assay for marker-assisted selection of soil-borne Rhizoctonia solani AG-2-2-IIIB resistance in sugar beet. Biology. 2021, 11(1), 49.
13. A.Y.M. Nevamea, L. Xiaa, Z. Wentinga, C.G. Nchongbohc, L. Wenhua, M.M. Hasand, ... & S. Longtinga. Validation of some disease-resistance molecular markers associated with multiple diseases in tomato for marker-assisted selection program. Scienceasia, 2020, 4, 19-29.
14. A. Mwamahonje, J.S.Y. Eleblu, K. Ofori, T. Feyissa, S. Deshpande, A.L. Garcia-Oliveira, R. Bohar, M. Kigoni, P. Tongoona.

Introgression of QTLs for Drought Tolerance into Farmers’ Preferred Sorghum Varieties. Agriculture 2021, 11, 883.

1. S. Chandran, B. Pukalenthy, K. Adhimoolam, D. Manickam, V. Sampathrajan, V. Chocklingam, ... & S. Natesan. Marker-assisted selection to pyramid the opaque-2 (o2) and β-carotene (crtRB1) genes in maize. Frontiers in genetics, 2019, 10, 859.