Chapter

Current Applicable DNA Markers for Marker Assisted Breeding in Rice (*Oryza sativa* L.)

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Abstract

Rice, (*Oryza sativa* L.) account as the second cereal most cultivated in the world. Unfortunately, global rice production is rendered by significant number abiotic and biotic stresses. Breeding for resistant variety through conventional breeding is an economical method; generally, it takes at least 10 years to release a new rice variety. Advance technology in molecular marker had revolutionized and irreversibly changes the disciplines of plant genetic and breeding. Integration of DNA-based markers in selection process enhances the effectiveness and accuracy of conventional plant breeding. It offers a novel tool for discovering and tagging alleles and genes specifically in plant. Ubiquitous of DNA marker-trait associations for diverse crops species are available with the findings of many quantitative trait loci (QTLs) mapping studies. The linkage drags, and time-consuming in conventional breeding can minimize with the application of DNA markers in plant breeding. The utilization of DNA marker in QTL mapping, MAS and gene pyramiding has been investigated. In this chapter, we discussed the recent utilizing markers in rice breeding program against abiotic and biotic stresses. In a few decades, molecular marker assisted breeding (MAB) provide a boundless task for breeders in attaining an important impact on crop development.

Keywords: abiotic stress, biotic stress, marker-assisted selection, rice, resistant

1. Introduction

Oryza sativa is an essential food crop contributing 30% of calories for at least 62.8% of the world population. Presently, rice was cultivated over 167.13 m ha within 114 countries with annual production of 486.62 mt (on milled basis) [1]. Majority of population in Asian consumed rice as a basic staple crop with over than 90% of rice were produced in Asia [2]. Revolution in Green technology played an important role in adoption of major increasing in rice production over last four decades. Approximately 450 million tons to 650 million tons of world rice consumption is projected to surge by 2011 and 2050, respectively [3].

Scientist and plant breeder are facing a new challenge in crop improvement due to climate change issue including limitation of water and cultivated land, new emerging pathogen and pest resulted serious yield losses. Among the major constraints, blast disease, bacterial blight, sheath blight, tungro viruses, brown spot and a number of grain discoloration diseases [4] accounted for serious rice losses.

Several amendments can be implemented to overcome these limitations such as breeding high yielding varieties, durable resistance to diseases and tolerance to abiotic stresses [5]. Rice yield potential can be enhanced by using numerous approaches such as conventional hybridization, ideotype breeding, heterosis breeding, wide hybridization and molecular breeding [6]. In molecular rice breeding, there are two main strategies can be used by applying the biotechnology concept which are MAS and Genetically Engineering. MAS like genetic engineering is a technique used to introgress targeted gene however its offers tools for specific selection for further breeding in existing plant material. The field of plant genetics and breeding has irreversibly changed with the development of DNA (or molecular markers) [7].

Since past two decade, revolutionized in molecular marker to enhance the effectiveness in breeding and to significantly shorten the development time of varieties bring about the concept of molecular marker assisted selection (MAS) as an efficient plant selection [8–10]. A genetic marker is any noticeable character or otherwise assayable phenotype, for which alleles at individual loci segregate in a Mendelian manner. The genetic markers covered include (1) morphological markers (2) biochemical markers (alloenzymes and other protein markers) and (3) molecular markers (based on DNA-DNA hybridization) [11, 12]. In this chapter, we present the latest DNA-based markers in a few studies related to abiotic and biotic stress genes using the MAS application in rice breeding programs. These markers have proven significantly useful from different findings published in screening, fine mapping and gene pyramiding approaches.

2. Molecular marker

Rediscovery Mendelian theory in the early part of 20th century, reveal that inheritance (genes) linked together in chromosome. Molecular marker defines as individual genes flanking within a defined close interval. It can be found at a specific location of the genome and linked with inheritance of a trait or gene [13]. Thottappilly et al. [14] refers molecular marker as polymorphism identified between species including proteins and nucleic acids. Collard et al. [15] had divided molecular marker into 3 major groups which are morphological marker, biochemical marker and DNA marker (**Figure 1**). However, Xu [16] had categorized markers into two

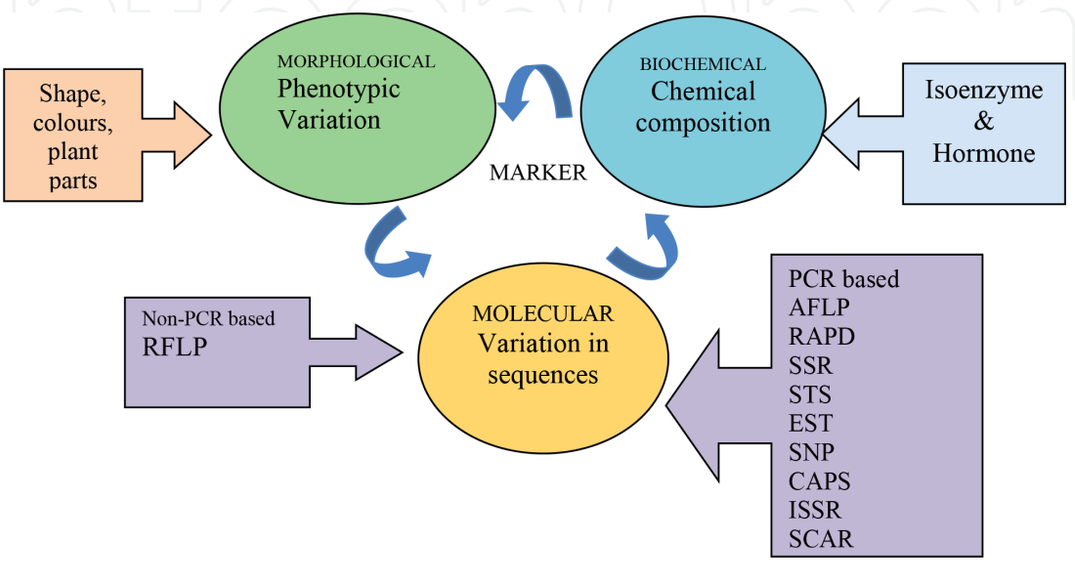

Figure 1.
Different types of molecular marker available.

broad groups based on detecting method; (i) classical marker and (ii) DNA marker. Classical marker was initiated with the mutations in the genomic loci controlling plant morphology [17] (**Table 1**). Advent in polymorphism detecting methods such as Polymerase Chain Reaction, Southern Blotting and Sequencing had invented the development of DNA markers (**Table 2**). Variation were detected at DNA level such as nucleotides changes ie; substitution (point) mutation, rearrangement (insertion and deletions) or errors in replication of tandemly repeated DNA [18].

Types of marker	**Morphological marker**	**Biochemical marker**
Advantages	Readily available	Highly reproducible
	Usually require only simple equipment	Applicable for measure a wide range of population genetic parameter
	The most direct measure of phenotype	Inexpensive
Disadvantages	Requires expertise on crop or species	Limited biochemical assay for detection
	Subjected to environmental influences	Phenotype-based analysis
	Limited in number	
Application	Conventional plant breeding program	Genetic diversity Map construction

Table 1.
Comparison between classical markers.

PCR based markers	**Advantages**	**Disadvantages**	**Application**
RFLP	Reliable and rigid	Harmful and tedious	Map construction Hybrid fixation Genetic diversity
	Easy to conduct across laboratory	Required a larger amount of DNA	
RAPD	A small amount of DNA required	Not transferable across laboratory	Genetic diversity Saturation mapping
	Simple, rapid and lowest cost	Irreproducibility	
	Single primer generates multiple loci		
SSR	Reliable, powerful and easy	Tedious and costly for initial establishment	Linkage and QTL mapping Marker-assisted selection Hybrids fixation
	Transferable between populations	Required polyacrylamide gel electrophoresis	
AFLP	Multiple loci	Required several DNA	Saturation mapping Genetic diversity
		Complicated	
SNP	Multiplexing large number of markers	Costly to assay	Fine mapping Map-based cloning
	Degraded DNA can be used	Marker has less allele	

Table 2.
Comparison of different PCR based marker.

Although there are many characteristics determine the suitable markers to be considered in MAS development; there are five main characteristics need to be considered [13, 19–22].

1. **Reliability:** Attribute of the desirable marker need to close proximity with the target loci, preferably less than 5 cm genetic distance or one marker every 10 cm [23].

2. **Repeatability and reproducible:** In plant breeding, a lot number of plants are usually screened for desirable marker pattern and result is required instantly. Therefore, level of simplicity in terms of time required and quick detection method are highly desirable for rapid selection. Statistical and bioinformatics software are significant for this purpose.

3. **Cost effective:** Marker assay should be not expensive feasible for selection process.

4. **Level of polymorphism:** Marker should discriminate between two similar accessions of species especially in parent breeding material.

5. **DNA quality and quantity:** A few markers require a complex DNA extraction technique and it too laboratories. In addition, it adds cost to the process.

3. Successful application of molecular marker in rice

Recently, MAB has become a well-known approach develop a superior genotype in rice. The aims of the breeding programs were to develop a rice variety with high yield potential, resistant to diseases and insects, tolerance to adverse environments, and acceptable grain quality. Since the first successful experiment stated for rice by Chen et al. [24] with introducing resistance to bacterial blight (BB) disease into Chinese hybrid parents. Till date, many varieties had been developed thru the application of molecular marker with backcrossing method. Varieties developed were improved from the wild type by incorporate desirable gene depending for biotic and abiotic stresses. Recently, in a few years there is a trend in producing more rice varieties that resistant in abiotic stress such as drought tolerance, submergence tolerance and salt tolerance. This demand comes from the periodic natural disaster in many regions of the rice producing country specifically in Asian country [25]. Resistant varieties against pathogenic disease were continuously developed. This scenario can be seen in blast disease where there are increasing number of resistant varieties were developed due to instability of the fungus pathogen.

Numerous molecular markers have been utilized in marker-assisted breeding however microsatellites are the preferable markers for plant breeding applications. The characteristic as codominant markers which inherited in a Mendelian fashion have emerged as a best choice compare to other markers. Additionally, microsatellite identify as preferable markers used in plant breeding programs due to ubiquitous, high polymorphism rates and wide range of distribution throughout the genome [26, 27]. Other markers possess major drawbacks including Restriction fragment length polymorphism (RFLP) markers which are not integrated with high-throughput technique; Random amplification of polymorphic DNA (RAPD) assays which are often not reproducible or immobile. AFLP analysis is not straightforward and commonly produces multiple fragments with the use of large genomic templates [28]. Application of molecular marker in rice breeding was demonstrated in **Table 3** to develop superior varieties in rice against abiotic and biotic stresses.

Stress	Traits	Gene/QTLs	Foreground and background selection		Reference
Abiotic	Deep roots	QTLs on chromosomes 1, 2, 7 and 9	RFLP and SSR	SSR	[29]
	Quality	waxy	RFLP	AFLP	[30]
	Root traits and aroma	QTLs on chromosomes 2, 7, 8, 9 and 11	RFLP and SSR	RFLP and SSR	[31]
	Submergence tolerance	*Sub1* QTL	Phenotyping and SSR	SSR	[32]
		Sub1 QTL	SSR	SSR	[33]
		*Sub1*QTL	SSR	STS	[34]
		Sub1 QTL	SSR	SSR	[35]
		*Sub*1	SSR	SSR	[36]
		Sub1	SSR	SSR	[37]
		Sub1	SSR	SSR	[38]
		Sub1	SSR	SSR	[39]
	Salt tolerance	*Saltol*	SSR	SSR	[40]
		Saltol QTL	SSR	SSR	[41]
		Saltol QTL	SSR	SSR	[42]
		Saltol QTL	SNP	SSR	[43]
	Early maturation	*Hd2*	SSR	SSR	[44]
	High yield and drought tolerance	*qDTY1.1, qDTY2.2, qDTY3.1, qDTY3.2, qDTY6.1*, and *qDTY12*	SSR	SSR	[45]
		qDTY1.1 + qDTY2.1 + qDTY3.1 + qDTY11.1 and two QTLs qDTY1.1 + qDTY11.1	SSR	SSR	[46]
	Heading time	QTL (*Hd1, Hd4, Hd5* and *Hd6*)	RFLP, STS, SSR, CAPS, dCAPS	RFLP, STS, SSR, CAPS, dCAPS	[47]
	Phosphorus tolerance	*Pup1*	SSR	SSR	[48]
	Drought tolerance	QTL	SSR	SSR	[42]
		MQTL1.1	SSR	SSR	[49]
		QTL	SSR	SSR	[50]

Stress	Traits	Gene/QTLs	Foreground and background selection		Reference
		(qDTY1.1, qDTY2.1)	SSR	SSR	[51]
		QTL	SSR	SSR	[52]
Biotic	Bacterial blight	*xa21*	STS	RFLP	[24]
		xa21	STS	AFLP	[53]
		xa5, xa13 and *xa21*	STS, CAPS	Not performed	[54]
		xa5, xa13 and *xa21*	STS	Not performed	[55]
		xa33	SSR	SSR	[56]
		xa34(t)	SSR	SSR	[57]
		xa35(t)	SSR	SSR	[58]
		xa38	SSR	SSR	[59]
		xa21	STS	STS	[60]
		xa5	CAPS	CAPS	[61]
		xa13, xa21	STS	SSR	[62]
		xa5 and *xa13*	CAPS	STS	[63]
		xa13 and *xa21*	CAPS	STS	[62, 64]
		xa13+ xa21 and *xa5 + xa21*	SSR	SSR	[65]
		xa13 and xa21	SSR	SSR	[66]
	Blast resistance	*Pi1, Pi2, Pi33* and *Pi54*	SSR	SSR	[67]
		Pi genes, *xa5*	SSR	SSR	[68]
		Pi54 and *Pi5*	SSR	SSR	[69]
		Pi1 and *Pi2*	SSR	SSR	[70]
		Pi1	SSR	ISSR	[71]
		Pikh and pi7(t)	SSR	SSR	[9]
		QTLs on chromosomes 1, 2, 11 and 12	SSR	SSR	[72]

Stress	Traits	Gene/QTLs	Foreground and background selection		Reference
	Submergence tolerance, disease resistance, quality	*Subchr9* QTL, *xa21*, *Bph* and blast QTLs and quality loci	SSR and STS	Not performed	[34]
	Thermo-sensitive genic male sterile (TGMS) and blast resistance	TGMS and *Pi* gene	SNP	SNP	[73]
	Brown planthopper resistance	*Bph14* and *Bph15*	InDEL	InDEL	[74]
		Bph18	SSR	SSR	[75]
	Gall midge resistance	*Gm8*	SSR	SSR	[76]
	Rice stripe resistance	*Stv-bi*	SSR	SSR	[77]

Table 3.
Application of molecular marker in MAB in rice.

4. Abiotic stresses

4.1 Deep root

Four QTL (chromosome 1, 2, 7 and 9) was introgressed to NIL developed from crossed between Azucena (donor parent) and IR64 (recipient parent) [29]. Twenty-nine NIL successfully developed in which three NIL carrying target 1 demonstrated significantly increased trait over IR64 while 3 out of 8 NIL consist combination target 1 and 7 exhibited higher root mass and depth. Four NIL carrying targets 9 showed increased maximum number of root length and 2 NIL showed no significant difference on root phenotypic compare to IR64. Results suggest the introgression of genes linked to root improvement is an ideal system compared to change the morphological characters of plant.

4.2 Quality

Waxy locus (*wx*-MH) from Mighui 63 was successfully introgressed into Zhenshan 97 which having poor agronomically quality including amylose content (AC), gel consistency (GC), gelatinization temperature (GT) and endosperm [30]. MABC scheme had been used to develop 3 backcross and 1 selfing generation. Improved variety of Minghui 63 (Shanyou 63) carrying *wx*-MH gene at 6.1 cm length have similar agronomy characteristic as Minghui 63 with low AC, high GC and GT with low grain opacity. Result indicated that waxy region gives significant effect on quality trait.

4.3 Root trait and aroma

MABC strategy had adopted by high yielding India upland rice variety, Kalinga III to insert 5 segments of QTL including 4 QTL carrying root length and thickness and 1 QTL control recessive aroma trait from Azucena donor variety which come from Philippines japonica rice [31]. This experiment was performed until 8 generation which using 3000 SSR markers for almost 8 years to develop NIL. Twenty-two NIL successfully develop which carrying QTL on root length at chromosome number 9 linked with RM242-RM201 and RM248 on chromosome 7 control for delayed flowering.

4.4 Submergence tolerance

Ten improves lines from OM 1490/IR 64 -Sub1 was developed from Marker-assisted backcrossing and having 90–99% survival under field submergence treatment [78]. Foreground selection was carried out using microsatellite, RM23805 marker which identified allele as 240 and 230 bp bands, respectively from OM1490, susceptible parent and IR64-*Sub*1, tolerant parent. Entire lines of OM1490/IR64-*Sub*1 introgression possess an increase rate of survival from original parent. In India, high yielding mega variety, Swarna had been incorporated with *Sub*1 gene by using three selections namely foreground, recombinant, and background selection, respectively. Findings indicated that superior variety Swarna successfully transformed into submergence tolerant variety within 2–3 years with three backcross generations [33]. Popular rice variety, AS996 successful incorporated *sub1* gene from rice variety IR64 as a donor parent through MABC. Insertion of *sub1* was confirmed with foreground SSR marker, ART5 and SC3 [36]. Background analysis was screened with 53 polymorphic SSR markers to assess BC_2F_1 and BC_3F_1 generations. The highest recurrent background was achieved 100% donor parent. Results suggest the development a new submergence tolerant rice variety

ASS996-SUB1 to stand with climate change. Superior varieties from India (Samba Mahsuri and CR1009), IRRI, Philippines (IR64), Laos (Thadokkham 1) and Bangladesh (BR11) successfully introgress *sub1* gene exhibited an improve survival rate compare to parental variety by applying MABC approach. Septiningsih et al. [79] studied the expression level of sub genes revealed that *sub1C*-1 was dominant over the *sub1A* allele however no reduction of expression observed when intolerant *sub1C* allele merged with the tolerant *sub1A*-1 allele. Survival rates of heterozygous plant with *sub1A* expression higher compared to homozygous tolerant parent. Existence of *sub*1A-2 will not significantly function in intolerant plant with turn off expression of sub*1C*-1. Study by Ahmad et al. [38] developed 30 newly lines of BC_2F_3 population from backcross of MR219 and Swarna-Sub1 with 95.37% recurrent parent genome (RPG) recovery. Rahman et al. [39] stated a marker-assisted backcross breeding (MABB) approach to incorporate the *Sub1* locus into popular rice varieties of southern India, CO 43, from tolerant FR13A variety. An elite NILs of CO 43 harboring *Sub1* locus possessing 95.78% of the recurrent parental CO 43 genome produced from genotyping and phenotyping analysis.

4.5 Salt tolerance

ASS996 a high yielding and widely grown cultivar in Vietnam had been cross with FL478, a highly salt tolerant rice variety using MABC strategy [40]. Four *Saltol*-AS996 introgression lines, QF3–1, QF3–2, QF 4–3-3, QF6–4 was developed with 3 foreground SSR markers, AP3206, RM3412 and RM10793 and 63 polymorphic markers were applied across rice genome for background analysis. The promising lines showed significant salt tolerance or similar compare to recurrent parent, FL478. MABC approach had been applied for introgressed *Saltol* QTL from donor parent, FL478 into Bacthom 7 recipient rice cultivar. Eighty-nine polymorphic SSR markers including 8 foreground markers to screen heterozygous plant carrying *Saltol* locus in each backcrossed generation. Background analysis demonstrated the restoration of recipient genome up to 96.8–100% in the best plant of BC_3F_1 generation. Finding concluded that improve version of salt tolerance-Bacthom 7 is relevant for cultivation in coastal areas of the Vietnamese Deltas [41]. De leon et al. [43] reported the utilization of SSR and SNP markers to characterize the ILs (Pokkali x 'Bengal) and identify 14 QTLs traits related to salinity tolerance.

4.6 Early maturation

In Indonesia, *Hd*2 gene located at chromosome 7 from Nipponbare was transferred to Code variety [44]. Two markers linked with early maturation trait, RM1362 and RM7601 was used to identify heterozygous carrying this gene in 2 backcross and 1 selfing generation. Improved lines showed a similar agronomic characteristic with code variety except for heading time which 73–85 days in improved version compared to recipient parents, 89 days. Maturation days for improved version of Code carrying *Hd*2 gene were 103–104 day which is at considerable maturation days. Result concluded the availability to use these improved lines in Indonesia for increasing rice production.

4.7 Heading time

Koshihikari and Kasalath were crossed by using MABC strategy to introgress 3 QTL segment which code for *Qdth*3, *Qdth*6 and *Qdth*8 [47]. Genotyping analysis was performed using five different types of DNA marker including RFLP, STS, SSR, CAPS and dCAPs. Three NIL successfully produced namely Wakei 367, Wakei 371 and Kanto IL1 which carrying Hd2 gene coded for heading time. All NIL exhibited

same morphological trait with Koshihikari with early heading time for Kanto IL1 (12 days), Wakei 367 (10 days) and Wakei 371 (11 days) compare to 95 days on Koshihikari.

4.8 Phosphorus tolerance

MABC approach had been successfully adapted to introgress a major quantitative trait locus (QTL), *Phosphorus uptake1* (*Pup1*), positioned on rice (*Oryza sativa*) chromosome 12 into two types of Indonesian rice varieties, irrigated and upland. SNP and CAPs had been used as foreground marker to identify desirable traits and background selection had been conducted using 47 to 61 polymorphic SSRs markers and restored 89–93% of the recipient genome in BC_2F_3 population. Phenotyping finding suggests that *Pup1* is effective to enhance grain yield under field treatment [48].

4.9 Drought tolerance

Crossing by hand emasculation and artificial pollination was performed between IR20 (*japonica*) and lowland (*indica*) cultivar to develop near isogenic lines (NILs) [42]. Foreground and background selection were conducted using 100 SSR markers that distributed entire genome. Five NILs exhibited high yield performance compare to IR20 in rain-fed and irrigated conditions. NIL-212 and 297 successfully incorporated with three and two root QTLs with NIL-297 demonstrated maximum value of nodal root and surface area whereas increase number of nodal roots than recurrent parent, IR20 observed in NIL 212. Backcrossed population (BC_2F_3) was developed through Marker Assisted Selection (MAS) to transfer QTLs for drought tolerant characters into Thai famous variety KMDL105 using 3 different donor parents, DH212, DH103 and DH126, respectively. Hundred and three KMDL 105 introgression lines were developed carrying selected target [80]. Genome analysis claimed that donor segment was found in non-carrier chromosomes and minimum proportion of recurrent genome was determined in carrier chromosome. Results suggest that background analysis is required to restore the recurrent genome segments. Sandhu et al. [46] enhance the grain yield of Samba Mahsuri under reproductive-stage drought (RS) through marker-assisted selection (MAS) and marker-assisted recurrent selection (MARS). Findings recorded four *qDTYs* ($qDTY_{1.1} + qDTY_{2.1} + qDTY_{3.1} + qDTY_{11.1}$) and two QTLs ($qDTY_{1.1} + qDTY_{11.1}$). Presence of gene MQTL1.1 responsible for the drought tolerance was identified from the cross (Sarjoo52 × Nagina- 22) in the study reported by Awasthi and Lal [49]. Ramchander et al. [50] study 101 Backcross Inbred Lines (BILs) of rice (BC_1F_5) for drought tolerance by develop a cross between Norungan//TKM9/Norungan. Marker aided selection used in this study efficiently pyramiding favorable QTL alleles to enhance drought tolerance in elite background of famous rice varieties using 15 polymorphic SSR markers. Muthu et al. [51] successfully developed abiotic stress-tolerant rice genotypes of the famous rice variety, Improved White Ponni (IWP) through a marker assisted backcross breeding approach by introgression major effect quantitative trait loci (QTLs) conferring tolerance against drought (*qDTY1.1*, *qDTY2.1*), salinity (*Saltol*), and submergence (*Sub1*). Backcrossed inbred lines (BILs) developed in this study showed minor effect caused by drought, salinity, and submergence. Root trait genes were introgressed through marker-assisted backcross breeding (MABC) between aerobic rice variety, AERON1 and MRQ74, a high yielding rice variety of Malaysia, [52]. Six best rice lines in BC_2F_1 with highest recovery percentage and resemble similar phenotypic appearance were identified with RM242 and RM263 foreground and 57 background markers.

4.10 Grain fragrance

Ahn et al. [81] was first discover the molecular marker linked with grain aroma. Lorieux [82] was used RFLP marker, RG28, which linked with aromatic located at chromosome 8 with genetic distance of 4.5 cm. The uniqueness of RG28 was interchangeable into STS marker and can be used to distinguish between fragrance and non-fragrance rice cultivars [83–85]. Improved version Pusa Bashmati and improved versions of PRR78 was successfully introgressed with quality trait from Basmati 370 as a donor with Manawthukha cultivar as a recipient using MABC scheme [86, 87].

4.11 Semi-dwarfing

*Sd*1 gene coded for semi-dwarf trait creates an interest among breeder as it enhances crop production. Advent in MAB had made it possible to track the introgression of *sd*1 gene by using SSR markers in *two traditional japonica varieties*, 'IR36' and 'Allorio' (IRGC284) [88].

5. Biotic stresses

5.1 Bacterial blight resistance

Marker assisted backcrossing coupled with stringent phenotypic screening was adapted to transfer BB resistance gene, *Xa*23 into Minghui63, YR293 and Y1671 which famous recipient lines. Foreground selection was screened with RM206 located at 1.9 cm near to the *Xa*23 locus [89]. The improved version of BB resistant was identical with original recipient for agronomic traits [30]. NSIC Rc142 and NSIC Rc154 donor cultivar for *Xa*4 + *Xa*5 + *Xa*21 genes were incorporated into susceptible cultivar IR64 in Philippines [61]. Currently, *Xa*13 and *Xa*21 had been successfully inserted into recurrent line PRR78 through MABC [90]. In Thailand, three backcrossed generation required to insert *Xa*21 (BB resistance gene) from IR1188 variety into famous KDML105 variety using MABC approach together with phenotypic selection. Similar agronomic characteristic was observed between developed introgressed lines having BB resistant gene (*Xa*21) with local variety KDML105 [91]. Foreground selection was screened with six SSR markers namely RM302, RM212, RM210, RM149, RM287, RM224 at different chromosome while 66 rice SSR markers was applied for background genotype. In China, popular restorer line, Yihui1577 was incorporated with *Xa*7 and *Xa*21 from Huahui20 thru MABC strategy [92]. Foreground marker were applied using RM20582 in existence of *Xa*7 to map a 0.14 cM interval at chromosome 6 between RM20582 and RM20593 [93]. Huang et al. [40] in his study successfully employed of *Xa*7, *Xa*21, *Xa*22 and *Xa*23 from Jinke 1 a cytoplasmic male sterile line into Huahui 1035 line using MABC. High yielding F_1 hybrid was used to develop more hybrid rice in China. Samba Mahsuri a mega high-yielding, fine grain-type variety was selected as donor for *Xa*21and *Xa*13 to incorporate into susceptible varieties, Taraori Basmati and Basmati 386 through MABC scheme [94]. Molecular backcross breeding was used to pyramided *Xa*21 + *xa*13 and *Xa*21 + *xa*5 combinations genes in a background of lowland cultivar, Jalmagna [65]. Twenty pyramided lines with targeted gene combinations showed resistant against BB pathogen. Balachiranjeevi et al. [66] reported the development of durable BB resistance through marker-assisted backcross breeding (MABB) into background of elite rice, DRR17B by introgress *Xa21* and *Xa33* dominant genes.

Six promising pyramiding lines of DRR17B with durable resistance and similar or superior agro-morphological attributes to DRR17B have been identified.

5.2 Blast resistance

C101A51 and Tetep contribute for *Piz*-5 and *Pi*54 blast gene was introgressed into PRR78 variety [69]. Two improved lines were developed, Pusa1602 (Piz5) and Pusa1603 (*Piz*54) through MABC breeding strategy by two backcross series. Foreground selection was performed by using AP5930 and RM206 while background analysis using SSR polymorphic markers revealed 89.01 and 87.88% in Pusa1602 and Pusa1603 lines recovery of RPG, respectively. Hybrid develop carrying blast resistance gene produced exhibited similar performance with parental Pusa RH10 in terms of yield, grain and cooking quality traits. Multiple resistance gene against leaf blast (*Pi*1), neck blast (*Pi*2) (donor BL122) and bacterial (*Xa*23) (donor CBB23) was successfully introgressed into Ronfeng B, a mega variety with early maturating line thru MABC scheme [70]. Improved lines D521, D524 was developed using three foreground markers (MRG4766, AP22 and RM206) and 131 polymorphic background markers. Proportional recipient genome for D521 and D524 were 96.18 and 96.56%, respectively after 4 backcrossed generations. The size of lesions exhibited from 0.77 to 1.18 cm with 96.7–100% resistance frequencies values. Rongfeng 3A was developed as an improve version of cytoplasmic male sterile line from Rongfeng by a series of conducive backcross breeding with *Pi*1, *Pi*2 and *Xa*23 gene. A series of superior lines with resistance to bacterial blight and blast diseases were developed by using MAB [30, 90, 95]. Hybrid Pusa RH10 is likely susceptible to BB and blast therefore by employing the bacterial and blast resistance gene, it will be improved adjustability to disease endemic parts and also retain the rice production. In Iranian, two crosses were made between susceptible, Tarom Mahali (TAM) and resistance cultivar, Khazar (KHZ) to increase blast resistance. Screening with 74 polymorphic markers on 192 $F_{2:3}$ families demonstrated both parental exhibited maximum genetic variation between the two varieties on entire chromosomes except chromosome 6, 7, 8 and 12 [96]. In Malaysia, improved version of MR219 was developed by introgression of *Pi-z* and *Pikh* and *Pi-b* by using two different donor, Pongsu Seribu 1 and 2 respectively [10, 97]. Hasan et al. [98] had successfully introgressed *Pi7(t)* and *Pikh* in MR264 from Pongsu Seribu 2 by using MABC approach. Thailand rice, the popular Sakon Nakhon (SKN) rice had been introgressed with QTLs on chromosomes 1, 2, 11 and 12 by using marker-assisted backcrossing. Srichant et al. [72] in his study presented 2 lines (SKN 39-10-19-29-12 and SKN 39-10-19-29-13) with high resistance to leaf and neck blast; and having similar agronomic traits as the SKN.

5.3 Brown plant hopper resistance

*Bph*3, gene coded for resistance against brown plant hopper had been introgressed into the famous Thai variety 'Khao Dawk Mali 105' [99]. Backcrossed population had been screened with RM589 and RM190, highly linked to *Bph*3 and *Wx*-RH loci. Fifty NIL were developed from background selection using 75 polymorphic SSR markers disperse 12 rice chromosomes. Cross had been made between Junambyeo, an elite japonica cultivar with IR65482-7-216-1-2 a donor indica rice by a series of backcrossing through MAB [75]. *Bph18* gene was transferred and 7312T4A, the most closely polymorphic STS marker was applied to screen the existence of the *Bph*18 gene in BPH-introgress backcrossed lines [100]. Two hundred and sixty SSR marker were used to screen the RPG percentage in the four NIL breeding lines.

5.4 Gall midge resistance

Gm8 resistance gene code for rice gall midge had been employed from donor parent Aganni into Samba Mashuri having g*ene Xa21* using MABC. Four plants identified in selfing population (BC2F2) carrying *Gm*8 and *Xa*21 genes and further tested for phenotypic selection against BB and gall midge [76]. Exploration of gm gene had been a great achievement in modern crop however resistant variety carrying 1 major gene easily lost the resistancy [101]. Till date, 11 Gm genes in the plant [102] and 7 pathogens have been identified [103].

6. Conclusion

Advent in marker technology has offer a remarkable progress in crop improvement specifically in rice. Present paper reviews the application of DNA markers in abiotic and biotic stresses in rice using MAS. Interval between molecular markers and gene/QTLs linked with target traits play a significant role in efficiency of MAS technique. Generally, marker will be validated in fine mapping study. Thus, the development of important marker associated with abiotic and biotic stresses resistance is proficient by QTL mapping experiments. MAB greatly increase the effectiveness and efficiency of breeding. Desirable individual in germplasm collection with genes or QTLs can be characterized in genotyping analysis by using the DNA markers for target genomic regions rather than phenotyping analysis. Pyramiding tolerance genes and QTLs will develop a resistant cultivar against abiotic and biotic stresses. MAS refers to selection by DNA markers linked to QTLs or target genes. Nowadays, DNA-based genetic markers play a significant part in the forthcoming of MAB and molecular genetics analysis for establishment of stress-tolerance and resistance in plants through molecular linkage maps. Knowledge of markers for abiotic and biotic tolerant traits presented here will serve a fundamental guide and help rice breeders in their MAB.

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References

[1] Shahbandeh M. World rice acreage 2010-2018. 2020. Available from: https://www.statista.com/statistics/271969/world-rice-acreage-since-2008/

[2] Bandumula N. Rice production in Asia: Key to global food security. Proceedings of the National Academy of Sciences, India Section B: Biological Sciences. 2017;**88**:1323-1328. DOI: 10.1007/s40011-017-0867-7

[3] Rejesus RM, Mohanty S, Balagtas JV. Forecasting Global Rice Consumption. Working Paper. North Carolina State University; 2012

[4] Singh PK, Nag A, Arya P, Kapoor R, Singh A, Jaswal R, et al. Prospects of understanding the molecular biology of disease resistance in Rice. International Journal of Molecular Sciences. 2018;*19*:1141

[5] Oladosu Y, Rafii MY, Samuel C, Fatai A, Magaji U, Kareem I, et al. Drought resistance in Rice from conventional to molecular breeding: A review. International Journal of Molecular Sciences. 2019;**20**:3519

[6] Khan MH, Dar ZA, Dar SA. Breeding strategies for improving Rice yield—A review. Agricultural Sciences. 2015;**6**:467-478. DOI: 10.4236/as.2015.65046

[7] Collard BCY, Mackill DJ. Marker-assisted selection: An approach for precision plant breeding in the twenty-first century. Philosophical Transactions of the Royal Society B. 2007;**363**:557-572

[8] Chukwu SC, Rafii MY, Ramlee SI, Ismail SI, Hasan MM, Oladosu YA, et al. Bacterial leaf blight resistance in rice: A review of conventional breeding to molecular approach. Molecular Biology Reports. 2019;**46**:1519-1532

[9] Hasan N, Rafii MY, Abdul Rahim H, Nusaibah SA, Mazlan N, Abdullah S. Genetic analysis and identification of SSR markers associated with rice blast disease in a BC_2 F_1 backcross population. Genetics and Molecular Research. 2015b;**16**(1):1-11

[10] Miah G, Rafii MY, Ismail MR, Puteh AB, Rahim HA, Asfaliza R, et al. Blast resistance in rice: A review of conventional breeding to molecular approaches. Molecular Reproduction and Development. 2013;**40**:2369-2388

[11] Nadeem MA, Nawaz MA, Shahid MQ, Doğan Y, Comertpay G, Yıldız M, et al. DNA molecular markers in plant breeding: Current status and recent advancements in genomic selection and genome editing. Biotechnology & Biotechnological Equipment. 2018;**32**(2):261-285

[12] Shabir G, Aslam K, Khan AR, Shahid M, Manzoor H, Noreen S, et al. Rice molecular markers and genetic mapping: Current status and prospects. Journal of Integrative Agriculture. 2017;**16**(9):1879-1891

[13] Jiang GL. Plant marker-assisted breeding and conventional breeding: Challenges and perspectives. Advances in Crop Science and Technology. 2013;**1**:105

[14] Thottappilly G, Magonouna HD, Omitogun OG. The use of DNA markers for rapid improvement of crops in Africa. African Crop Science Journal. 2000;**8**(1):99-108

[15] Collard BCY, Jahufer MZZ, Brouwer JB, Pang ECK. An introduction to markers, quantitative trait loci (QTL) mapping and marker-assisted selection for crop improvement: The basic concepts. Euphytica. 2005;**142**:169-196

[16] Xu Y. Molecular Plant Breeding. Wallington, UK: CABI International; 2010. p. 734. ISBN 978-184593-392-0

[17] Xu Y, Crouch JH. Marker-assisted selection in plant breeding: From publications to practice. Crop Science. 2008;**48**:391-407

[18] Modi D, Pandey B. Potential of molecular markers in plant biotechnology. Plant Omics. 2009;**2**:141-162

[19] Farooq S, Azam F. Molecular markers in plant breeding-II. Some pre-requisites for use. Pakistan Journal of Biological Sciences. 2002;**5**:1141-1147

[20] Mackill DJ, Ni J. Molecular mapping and marker assisted selection for major-gene traits in rice. In: Khush GS, Brar DS, Hardy B, editors. Proceeding Fourth International Rice Genetics Symposium. Los Baños, The Philippines: International Rice Research Institute; 2000. pp. 137-151

[21] Mohler V, Singrun C. General considerations: Marker-assisted selection in biotechnology in agriculture and forestry. Molecular Marker Systems. 2004;**55**:305-317

[22] Sabina A, Shafiq MA, Wani A, Bhat KA, Chalkoo S, Mir MR, et al. Marker assisted selection in rice. Journal of Phytopathology. 2010;**2**(10):66-81

[23] Boopathi NM. Marker-assisted selection (MAS). In: Genetic Mapping and Marker Assisted Selection. Singapore: Springer; 2020

[24] Chen S, Lin XH, Xu CG, Zhang QF. Improvement of bacterial blight resistance of 'Minghui 63', an elite restorer line of hybrid rice, by molecular marker-assisted selection. Crop Science. 2000;**40**:239-244

[25] Aryal JP, Sapkota TB, Khurana R, Chhetri AK, Rahut DB, Jat ML. Climate change and agriculture in South Asia: Adaptation options in smallholder production systems. Environment, Development and Sustainability. 2019;**21**:1-31. DOI: 10.1007/s10668-019-00414-4

[26] Kalia RK, Rai MK, Kalia S, Dhawan KD. Microsatellite markers: An overview of the recent progress in plants. Euphytica. 2011, 2011;**177**(3):309-334. DOI: 10.1007/s10681-010-0286-9

[27] Morgante M, Hanafey H, Powell W. Microsatellites are preferentially associated with nonrepetitive DNA in plant genome. Nature Genetics. 2002;**30**(22):194-200

[28] Paun O, Schönswetter P. Amplified fragment length polymorphism (AFLP) – An invaluable fingerprinting technique for genomic, transcriptomic and epigenetic studies. Methods in Molecular Biology. 2012;**862**:75-87

[29] Shen ZK, Wang M, Li Y, Jackson D, Yin A, Dong D, et al. Crustal deformation associated with the Altyn Tagh fault system, western China, from GPS. Journal of Geophysical Research. 2001;**106**(30):607-621

[30] Zhou WC, Kolb FL, Bai GH, Domier LL, Boze LK, Smith NJ. Validation of a major QTL for scab resistance with SSR markers and use of marker-assisted selection in wheat. Plant Breeding. 2003;**122**:40-46

[31] Steele KA, Price AH, Shashidhar HE, Witcombe JR. Marker-assisted selection to introgress of rice QTLs controlling root traits and aroma into an Indian upland rice variety. Theoretical Application Genetics. 2006;**112**:208-221

[32] Mackill DJ, Collard BCY, Neeraja CN, Rodriguez RM, Heuer S, Ismail AM. QTLs in rice breeding: Examples for abiotic stresses. In: Brar DS, Mackill DJ, Hardy B, editors. Rice Genetics 5: Proceedings of the International Rice Genetics Symposium. Manila: International Rice Research Institute; 2006. pp. 155-167

[33] Neeraja C, Maghirang-Rodriguez R, Pamplona A, Heuer S, Collard B,

Septiningsih E. A marker-assisted backcross approach for developing submergence-tolerant rice cultivars. Theoretical and Applied Genetics. 2007;**115**:767-776

[34] Toojinda T, Tragoonrung S, Vanavichit A, Siangliw JL, Pa-In N, Jantaboon J, et al. Molecular breeding for rainfed lowland rice in the Mekong region. Plant Production Science. 2005;**8**:330-333

[35] Iftekharuddaula M, Ahmed HU, Ghosal S, Moni ZR, Md SA. Development of new submergence tolerant Rice variety for Bangladesh using marker-assisted backcrossing. Rice Science. 2015;**22**(1):201

[36] Cuc LM, Huyen LTN, Hien PTM, Hang VTT, Dam NQ, Mui PT, et al. Application of marker assisted backcrossing to introgress the submergence tolerance QTL *Sub*1 into the Vietnam elite rice variety-AS996. American Journal of Plant Sciences. 2012;**3**:528-536

[37] Ling ZZ, Mew T, Wang JL, Lei CL, Huang L. Development of Chinese near-isogenic lines of rice and their differentiating ability to pathogenic races of *Pyricularia grisea*. Science in Agriculture. 2000;**33**:1-8

[38] Ahmed F, Rafii MY, Ismail MR, Juraimi AS, Rahim HA, Latif MD, et al. The addition of submergence-tolerant Sub1 gene into high yielding MR219 rice variety and analysis of its BC2F3 population in terms of yield and yield contributing characters to select advance lines as a variety. Biotechnology & Biotechnological Equipment. 2016;**30**(5):853-863. DOI: 10.1080/13102818.2016.1192959

[39] Rahman H, Dakshinamurthi V, Ramasamy S, Manickam S, Kaliyaperumal AK, Raha S, et al. Introgression of submergence tolerance into CO 43, a popular Rice variety of India, through marker-assisted backcross breeding. Czech Journal of Genetics and Plant Breeding. 2018;**54**(3):101-108

[40] Huyen LTN, Cuc LM, Ismail AM, Ham LH. Introgression the salinity tolerance QTLs *Saltol* into AS996, the elite rice variety of Vietnam. American Journal of Plant Sciences. 2012;**3**:981-987

[41] Vu HTT, Le DD, Ismail AM, Le HH. Marker-assisted backcrossing (MABC) for improved salinity tolerance in rice (Oryza sativa L.) to cope with climate change in Vietnam. Australian Journal of Crop Science. 2012;**6**(12):1649-1654

[42] Suji KK, Prince KSJ, Mankhar PS, Kanagaraj P, Poornima R, Amutha K, et al. Evaluation of rice (*Oryza sativa* L.) near iso-genic lines with root QTLs for plant production and root traits in rainfed target populations of environment. Field Crops Research. 2012;**137**:89-96

[43] De Leon TB, Linscombe S, Subudhi PK. Identification and validation of QTLs for seedling salinity tolerance in introgression lines of a salt tolerant rice landrace 'Pokkali'. PLoS One. 2017;**12**(4):e0175361. DOI: 10.1371/journal.pone.0175361

[44] Fatimah MA, Emmy FA, Kusairi M, Muhammad T. Food security: Self-sufficient of rice in Malaysia. International Journal of Management. 2011;**18**(2):83-100

[45] Kumar A, Dixit S, Ram T, Yadaw RB, Mishra KK, Mandal NP. Breeding high-yielding drought-tolerant rice: Genetic variations and conventional and molecular approaches. Journal of Experimental Botany. 2014;**65**:6265-6278. DOI: 10.1093/jxb/eru363

[46] Sandhu N, Dixit S, Swamy BPM, Vikram P, Venkateshwarlu C, Catolos M, et al. Positive interactions of major-effect

QTLs with genetic background that enhances rice yield under drought. Scientific Reports. 2018;**8**:1626. DOI: 10.1038/s41598-018-20116-7

[47] Takeuchi N, Ebitani T, Yamamato T, Sato H, Ohta H, Nemoto H, et al. Development of isogenics of rice cultivar Koshihikari with early and late heading by marker-assisted selection. Breeding Science. 2006;**56**:405-413

[48] Chin JH, Gamuyao R, Dalid C, Bustamam M, Prasetiyono J, Moeljopawiro S, et al. Developing rice with high yield under phosphorus deficiency: Pup1 sequence to application. Plant Physiology. 2011;**156**:1202-1216

[49] Awasthi S, Lal JP. Application of molecular markers to improve drought tolerance and yield enhancement in upland rice. Annals of plant sciences. Annals of Plant Sciences. 2015;**4**(04):1051-1056

[50] Ramchander S, Raveendran M, Robin S. Mapping QTLs for physiological traits associated with drought tolerance in rice (Oryza Sativa L.). Journal of Investigative Genomics. 2016;**3**(3):56-61. DOI: 10.15406/jig.2016.03.00052

[51] Muthu V, Abbai R, Nallathambi J, Rahman H, Ramasamy S, Kambale R, et al. Pyramiding QTLs controlling tolerance against drought, salinity, and submergence in rice through marker assisted breeding. PLoS One. 2020;**15**(1):e0227421. DOI: 10.1371/journal.pone.0227421

[52] Jalil MA, Juraimi AS, Yusop MR, Uddin MK, Hakim MA. Introgression of root trait genes for drought tolerance to a Malaysian rice variety by marker-assisted backcross breeding. International Journal of Agriculture and Biology. 2018;**20**:119-126

[53] Chen S, Xu CJ, Lin XH, Zhang QQ. Improving bacterial blight resistance of '6078', an elite restorer line of hybrid rice, by molecular marker-assisted selection. Plant Breeding. 2001;**120**(2):133-137

[54] Sanchez AC, Brar DS, Huang N, Li Z, Khush GS. Sequence tagged site marker-assisted selection for three bacterial blight resistance genes in rice. Crop Science. 2000;**40**:792-797

[55] Singh S, Sidhu JS, Huang N, Vikal Y, Li Z, Brar DS, et al. Pyramiding three bacterial blight resistance genes (*xa5*, *xa13* and *Xa21*) using marker-assisted selection into *indica* rice cultivar PR106. Theoretical and Applied Genetics. 2001;**102**:1011-1015

[56] Natrajkumar P, Sujatha K, Laha GS, Srinivasarao K, Mishra B, Viraktamath BC, et al. Identification and fine mapping of *Xa33*, a novel gene for resistance to *Xanthomonas oryzae* pv. *oryzae*. Phytopathology. 2012;**102**:222-228

[57] Chen S, Liu X, Zeng L, Ouyang D, Yang J, Zhu X. Genetic analysis and molecular mapping of a novel recessive gene xa34(t) for resistance against *Xanthomonas oryzae* pv. Oryzae. Theoretical and Applied Genetics. 2011;**122**:1331-1338

[58] Guo S, Zhang D, Lin X. Identification and mapping of a novel bacterial blight resistance gene Xa35(t) originated from Oryza minuta. Scientia Agricultura Sinica. 2010;**43**:2611-2618

[59] Bhasin H, Bhatia D, Raghuvanshi S, Lore JS, Sahi GK, Kaur B, et al. New PCR-based sequence-tagged site marker for bacterial blight resistance gene Xa38 of rice. Molecular Breeding. 2012;**30**:607-611

[60] Ronald PC, Albano B, Tabien R, Abenes L, Wu KS, McCouch S, et al. Genetic and physical analysis of the rice bacterial blight disease resistance locus, Xa27. Gene. 1992;**236**:113-120

[61] Toenniessen GH, O'Toole JC, DeVries J. Advances in plant biotechnology and its adoption in developing countries. Current Opinion in Plant Biology. 2003;**6**:191-198

[62] Joseph M, Gopalakrishnan S, Sharma RK, Singh VP, Singh AK, Mohapatra T. Combining bacterial blight resistance and basmati quality characteristics by phenotypic and molecular marker-assisted selection in rice. Molecular Breeding. 2004;**13**:377-387

[63] Sundaram RM, Priya MRV, Laha GS, Rani NS, Rao PS, Balachandran SM, et al. Introduction of bacterial blight resistance into Triguna, a high yielding, mid-early duration rice variety by molecular marker assisted breeding. Biotechnology Journal. 2009;**4**:400-407

[64] Gopalkrishnan RK, Rajkumar AK, Josheph M, Singh VP, Singh AK, Bhat KV, et al. Integrating marker assisted background analysis with foreground selection for identification of superior bacterial blight resistant recombinants in basmati rice. Plant Breeding. 2008;**127**:131-139

[65] Pradhan SK, Pandit E, Pawar S, et al. Development of flash-flood tolerant and durable bacterial blight resistant versions of mega rice variety 'Swarna' through marker-assisted backcross breeding. Scientific Reports. 2019;**9**:12810. DOI: 10.1038/s41598-019-49176-z

[66] Balachiranjeevi CH, Bhaskar NS, Abhilash KV, Harika G, Mahadev Swamy HK, Sk H, et al. Marker-assisted pyramiding of two major, broad-spectrum bacterial blight resistance genes, *Xa*21 and *Xa*33 into an elite maintainer line of rice, DRR17B. PLoS One. 2018;**13**(10):e0201271

[67] Divya B, Robin S, Rabindran R, Senthil S, Raveendran M, John Joel A. Marker assisted backcross breeding approach to improve blast resistance in Indian rice (*Oryza sativa*) variety ADT43. Euphytica. 2014;**200**:61-77

[68] Pinta W, Toojinda T, Thummabenjapone P, Sanitchon J. Pyramiding of blast and bacterial leaf blight resistance genes into rice cultivar RD6 using marker assisted selection. African Journal of Biotechnology. 2013;**12**(28):4432-4438

[69] Singh VK, Singh A, Singh SP, Ellur RK, Choudhary V, Sarkel S, et al. Incorporation of blast resistance into "PRR78", an elite basmati rice restorer line, through marker assisted backcross breeding. Field Crops Research. 2012;**128**:8-16

[70] Fu CY, Wu T, Liu WG, Wang F, Li JH, Zhu XY, et al. Genetic improvement of resistance to blast and bacterial blight of the elite maintainer line Rongfeng B in hybrid rice (*Oryza sativa* L.) by using marker-assisted selection. African Journal of Biotechnology. 2012;**11**:13104-13124. DOI: 10.5897/AJB12.1465

[71] Liu DW, Oard SV, Oard JH. High transgene expression levels in sugarcane (*Saccharum officinarurn* L) driven by rice ubiquitin promoter RUBQ2. Plant Science. 2003;**165**:743-750

[72] Srichant N, Chankaew S, Monkham T, Thammabenjapone P, Sanitchon J. Development of Sakon Nakhon rice variety for blast resistance through marker assisted backcross breeding. Agronomy. 2019;**9**(2):67

[73] Jiang J, Yang D, Ali J, Mou T. Molecular marker-assisted pyramiding of broad-spectrum disease resistance genes, *Pi2* and *Xa23*, into GZ63-4S, an elite thermo-sensitive genic male-sterile line in rice. Molecular Breeding. 2015;**35**:1-12

[74] Yu K, Park SJ, Poysa V. Marker-assisted selection of common beans for

resistance to common bacterial blight: Efficacy and economics. Plant Breeding. 2000;**119**:411-415

[75] Suh JP, Yang SJ, Jeung JU, Pamplona A, Kim JJ, Lee JH, et al. Development of elite breeding lines conferring Bph18 gene-derived resistance to brown plant hopper (BPH) by marker-assisted selection and genome-wide background analysis in japonica rice (*Oryza sativa* L.). Field Crops Research. 2011;**120**:215-222

[76] Sama VSAK, Himabindu K, Naik SB, Sundaram RM, Viraktamath BC, Bentur JS. Mapping and marker-assisted breeding of a gene allelic to the major Asian rice gall midge resistance gene *Gm8*. Euphytica. 2012;**187**:393-400

[77] Xu J. Pyramiding of two BPH resistance genes and *Stv-bi* gene using marker-assisted selection in japonica rice. Crop Breeding and Applied Biotechnology. 2013;**13**:99-106

[78] Lang NT, Phuoc NT, Ha PTT, Toan TB, Buu BC, Reinke R, et al. Development of submergence tolerant breeding lines for Vietnam. Sabrao Journal of Breeding and Genetics. 2015;**47**:448-459

[79] Septiningsih EM, Pamplona AM, Sanchez DL, Neeraja CN, Vergara GV, Heuer S, et al. Development of submergence-tolerant rice cultivars: The Sub1 locus and beyond. Annals of Botany. 2008;**103**:151-160

[80] Siangliw JL, Jongdee B, Pantuwan G, Toojinda T. Developing KDML105 backcross introgression lines using marker-assisted selection for QTLs associated with drought tolerance in rice. Science Asia. 2007;**33**:207-214

[81] Ahn SN, Bollich CN, Tanksley SD. RFLP tagging of a gene for aroma in rice. Theoretical Application Genetic. 1992;**84**:825-828

[82] Lorieux M, Petrov M, Huang N, Guiderdoni E, Ghesquière A. Aroma in rice: Genetic analysis of a quantitative trait. Theoritical Application Genetic. 1996;**93**:1145-1151

[83] Lang NT, Buu BC. Initial marker-assisted selection in rice breeding at Cuu Long Delta Rice Research Institute. Omonrice. 2010;**17**:8-21

[84] Jin Q, Waters D, Cordeiro GM, Henry RJ, Reinke RF. A single nucleotide polymorphism (SNP) marker linked to the fragrance gene in rice (*Oryza sativa* L.). Plant Science. 2003;**165**:359-364

[85] Mahdav MS, Pandey MK, Kumar PR, Sundaram RM, Prasad GSV, Sudarshan L, et al. Identification and mapping of tightly linked SSR marker for aroma trait for use in marker assisted selection in rice. Rice Genetics Newsletter. 2009;**25**:38-39

[86] Yi M, Nwe KT, Vanavichit A, Chai-arree W, Toojinda T. Markerassisted backcross breeding to improve cooking quality traits in Myanmar ricecultivar Manawthukha. Field Crops Residue. 2009;**113**:178-186

[87] Singh AK, Gopalakrishnan S, Singh VP, Prabhu KV, Mohapatra T, Singh NK, et al. Marker assisted selection: A paradigm shift in Basmati breeding. Indian Journal Genetic Plant Breeding. 2011;**71**(2):120-128

[88] Negrão S, Palaniappan J, Maroco J, Lourenço T, Mackill D, Oliveira MM. Bridging sd1 molecular knowledge with recent breeding strategies for the improvement of traditional rice varieties: A japonica case-study. African Journal Biotechnology. 2010;**9**:2192-2200

[89] Pan HJ, Wang CL, Zhao KJ, Zhang Q, Fan YL, Zhou SC, et al. Molecular mapping by PCR-based markers and marker-assisted selection of Xa23 a bacterial blight resistance

gene in rice. Acta Agronomica Sinica. 2003;**29**(4):501-507

[90] Basavaraj SH, Singh VK, Singh A, Singh A, Singh A, Yadav S, et al. Marker-assisted improvement of bacterial blight resistance in parental lines of Pusa RH10, a superfine grain aromatic rice hybrid. Molecular Breeding. 2010;**26**:293-305

[91] Win KM, Korinsak S, Jantaboon J, Siangliw M, Lanceras-Siangliw J, Sirithunya P, et al. Breeding the Thai jasmine rice variety KDML105 for non-age-related broad-spectrum resistance to bacterial blight disease based on combined marker-assisted and phenotypic selection. Field Crops Residue. 2012;**137**:186-194

[92] Xu J, Jiang J, Dong X, Ali J, Mou T. Introgression of bacterial blight (BB) resistance genes Xa7 and Xa21 into popular restorer line and their hybrids by molecular marker-assisted backcross (MABC) selection scheme. African Journal of Biotechnology. 2012;**11**:8225-8233

[93] Chen S, Huang Z, Zeng L, Yang J, Liu Q, Zhu X. High-resolution mapping and gene prediction of *Xanthomonas Oryzae* pv. Oryzae resistance gene Xa7. Molecular Breeding. 2008;**22**:433-441. DOI: 10.1007/s11032-008-9187-1

[94] Pandey MK, Shobharani N, Sundaram RM, Laha GS, Madhav MS, Rao KS, et al. Improvement of two traditional Basmati rice varieties for bacterial blight resistance and plant stature through morphological and marker-assisted selection. Molecular Breeding. 2013;**31**:239-246

[95] Basavaraj SH, Singh VK, Atul S, Devender S, Nagarajan M, Mohapatra T, et al. Marker aided improvement of Pusa 6B, the maintainer parent of rice hybrid Pusa RH10, for resistant to bacterial blight. Indian Journal of Genetics. 2009;**69**(1):10-16

[96] Sabouri H, Sabouri A, Jafarzadeh MR, Mollashahi M. Detection of QTLs controlling field blast resistance in rice (*Oryza sative* L.). Plant Omics. 2011;**4**:1-5

[97] Tanweer FA, Rafii MY, Sijam K, Rahim HA, Ahmed F, Ashkani S, et al. Introgression of Blast Resistance Genes (Putative Pi-b and Pi-kh) into Elite Rice Cultivar MR219 through Marker-Assisted Selection. Frontier Plant Science. 2015;**6**:1002. DOI: 10.3389/fpls.2015.01002

[98] Hasan MM, Rafii MY, Ismail MR, Mahmood M, Rahim HA, Alam MA. Marker-assisted backcrossing: A useful method for rice improvement. Biotechnology Equipment. 2015a;**29**:237-254

[99] Jairin J, Teangdeerith S, Leelagud P, Kothcharerk J, Sansen K, Yi M, et al. Development of rice introgression lines with brown planthopper resistance and KDML105 grain quality characteristics through marker-assisted selection. Field Crop Residue. 2009;**110**:263-271

[100] Jena KK, Jeung JU, Lee JH, Choi HC, Brar DS. High-resolution mapping of a new brown planthopper (BPH) resistance gene, Bph18(t), and marker-assisted selection for BPH resistance in rice (*Oryza sativa* L.). Theoretical Application Genetics. 2006;**112**:288-297

[101] Bentur JS, Srinivasan TE, Kalode MB. Occurrence of a virulent gall midge (GM), Orseolia oryzae Wood-Mason biotype in Andhra Pradesh, India. International Rice Research Newsletter. 1987;**12**(3):33-34

[102] Himabindu K, Suneetha K, Sama VSAK, Bentur JS. A new rice gall midge resistance gene in the breeding line CR57-MR1523, mapping with flanking markers and development of NILs. Euphytica. 2010;**174**:179-187

[103] Vijaya LP, Amudhan S, Himabindu K, Cheralu C, Bentur JS. A new biotype of the Asian rice gall midge Orseolia oryzae (Diptera: Cecidomyiidae) characterized from the Warangal population in Andhra Pradesh, India. International Journal Insect Science. 2006;**26**:207-211