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Applications of molecular markers for bacterial blight resistant varieties in rice (*Oryza sativa* L.)

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Abstract: Bacterial blight is one of devasting disease in almost all rice growing countries. The most effective, economic and environmental strategy for control of this disease is to develop rice varieties with genetic resistance. However, new pathotype has overcome single gene for resistance in the new cultivars. So, plant breeders are concentrating to develop high yielding varieties with durable resistance using novel technologies. Molecular marker technology has progressed tremendously in the past decade for genetic improvement of field crops. Molecular markers can improve efficiency of breeding in different ways for trait in segregating population like identify plants with target gene in maximum recovery portion of recurrent parent. The transfer of two or three genes into single variety with the help of molecular marker is expected to lead to more durable resistance. Thus, thus review describes progress made in the development of bacterial blight resistance rice varieties using Marker Assisted Selection.

Keywords: Bacterial blight, Durable resistance, Gene pyramiding, Molecular markers, Rice

INTRODUCTION

Rice plays an essential role to human population by providing nutrients, vitamins and minerals in terms of growth and development. India is second largest producer (106.54 metric tonnes) with capacity of yield (2424 kg/ha) (Ministry of Agricultural and Cooperation, India, 2014) of rice next to china. According to UN human population survey, Indian population is expected to reach 1.7 billion in 2050 (World Population Prospects, 2015). This population statistics clearly indicated that the productivity of crop varieties has to be increased to feed over increasing population. Bacterial blight (causes by *Xanthomonas oryzae* pv. *oryzae*) is one of destructive disease of rice that causes as much as 80% yield reduction (Pradhan *et al*., 2015, Arunakumari *et al*., 2016).

Bacterial blight is a vascular disease resulting in tannish gray to white lesions along the veins. The disease first appears in the seed bed as tiny, water soaked spots at the margin of fully developed lower leaves. As the spots enlarge, the leaves turn yellow, dry rapidly and wither. On leaf blades, lesions usually begin at the margin, a few are formed at the tip as water soaked stripes. The lesions enlarge both in length and width has a wavy margin and turns yellow within a few days. As the disease advances the lesions cover the entire blade, turn white and later become grayish colour. The entire blade may soon become involved and gets dried up. This disease occurs at all growth stages, but it is common from maximum tillering until maturity. In severely diseased fields, the infection may reach the grains (Mehrotra and Agarwal, 2003).

In earlier days for disease resistance breeding, breeders were used to inject spores into planting materials. After two/three weeks, evaluations were taken in those materials for disease development based on the lesions on the leaves. But the pathogen cause disease in experimental materials only under favorable conditions otherwise even susceptible plant would behave like resistant plant (Charpe *et al.,* 2012). So, breeders need new technique to evaluate the plant materials in the absence of pathogen/pest environment. As a result, molecular marker technology was developed by various workers in 21st century.

Era of molecular markers: In crop plants, first Linkage RFLP (Restriction Fragment Length Polymorphism) map was developed in Tomato (Bernatzky and Tanksley, 1986) and it was gradually extended into other crop plant also. To overcome the limitation of RFLP (requirement of quantity of DNA is high and use of radioactive probes); numerous DNA marker system based on Polymerase Chain Reaction (PCR) were developed *viz.,* RAPD (Random Amplified Polymorphic DNA) by Williams *et al*., 1990; AFLP (Amplified Fragment Length Polymorphism) was developed by Vos *et al*., 1995 and SSR (Simple Sequence Repeat) markers.

Application of molecular markers to plant breeders: Marker Assisted Back crossing strategies involves indirect selection process *viz.,* Marker Assisted Foreground Selection (target gene) and Marker As-

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Hierarchy in molecular marker evolution:

Restriction Fragment Length Polymorphism (RFLP)	In Initial days, various rice group scientists had developed RFLP linkage map (McCouch *et al*., 1988 and Causse *et al*., 1994) for molecular analysis in rice. **Disadvantage:** a. RFLP requires large quantities of good quality DNA. b. RFLP analysis is labor intensive and time consuming (Lateef, 2015)
Polymerase Chain Reaction (PCR) markers	In the year 1983, Kary Mullis discovered PCR to copy or amplify specific DNA sequence of living organism. In initial days, klenow fragment was used for amplification in PCR but the drawback of this enzyme operates at optimum reaction temperature at 37^{O} C. Because of heat liability, technologist have to add fresh enzyme each and every cycle as a result it leads to time consuming and labor intensive (Oste, 1989) But report of *Thermus acquaticus* enzyme by Saiki *et al*., (1988) which operates at extreme heat conditions (72^{O}C) makes PCR technology into next level. Now a day, there are numerous PCR based molecular markers is available for use in molecular breeding *viz*., Random Amplified Polymorphic DNA (RAPD), Amplified Fragment Length Polymorphisms (AFLP), Sequence-Tagged Site (STS), Expressed Sequence Tag (EST), sequence-characterized amplified regions (SCAR), Inter-Simple Sequence Repeats (ISSR), Cleaved Amplified Polymorphic Sequences (CAPS). Each and every marker system has its own advantages and disadvantages. Among different marker system, Simple Sequence Repeat is mostly commonly used marker because of its simplicity, co dominant marker *i.e.* able to differentiate between homozygous and heterozygous plant in segregating populations, friendly to use and reproducible result across all over the world (Kumar *et al*., 2009; Singh and Sengar, 2015; Wijerathna, 2015). Availability of SSR linkage map in rice was developed by Temnykh *et al*., (2001) and Orjuela *et al*. (2010) make possible to tag the individual gene for the concerned trait. It facilitates the selection of desired plants with the target gene even in the absence of pathogen/pest environment (Sanchez *et al*., 2000).
Single-Nucleotide Polymorphism (SNP)	SNP is a variation in a single nucleotide at the same locus in the genome between two individuals of the same species. The variation between two individual it's because of addition/deletion/ substitution of bases. The number of markers is highly abundant as compared to other markers system as a result polymorphism rate is high; detection and assay of SNP automated allows large scale genotyping at a short time (Mammadov *et al*., 2012). The details of SNP marker of rice are available in *OryzaSNP* and it needs minimum quantity of DNA, automation of genotype and largest amount of DNA polymorphism (Huq *et al*., 2016; kurokawa *et al*., 2016). Newly developed SNP markers are amenable to validation and once it is validated, it will be useful in molecular breeding approaches (Agarwal *et al*., 2016)

sisted Background Selection (proportion of recurrent parent genome in desirable plant). In Marker Assisted Foreground Selection, target genes are transferred into recipient variety with the help of flanking markers without any linkage drag (Shanthi *et al*., 2010).

The selection of plant with target gene in segregating population is carried out in nursery stage even in the absence of pathogen/pest environment. So, only the desirable plants will be transplanted to main field and it saves like land, water and energy resources (Basavaraj *et al*., 2010 and Sanchez *et al.,* 2000). The main objective of Marker Assisted Background Selection is to improve/replace defective gene of recurrent parent from donor parent with the maximum portion of recurrent parent genome. Parental polymorphism analysis has been carried out between recurrent parent and donor parent using markers selected from each of the 12 linkage map of Temnykh *et al.,* (2001) and Universal Core Genetic Map (Orjuela *et al*., 2010). Polymorphic SSR markers identified during parental polymorphism studies will used to identify plants with maximum recovery of recurrent parents in selected plants of backcross segregation populations**.** Marker assisted selection speed up the breeding process and shorten the development time of varieties (Cuc *et al*., 2012; Toledo *et al*., 2015)

Gene pyramiding: Generally, disease resistant variety have single gene for resistance and the durability of such gene is no longer for resistance. Since continuous pressure exert on the pathogen to develop virulence against resistant gene either by means of mutation or recombination. So, to develop long lasting resistant variety the concept of Gene pyramiding came into existences *i.e.* transfer of one or more genes into single variety. Because of Quantitative 'Complementation' (Basavaraj *et al.,* 2010) or 'synergistic action' (Zhang *et al.,* 1996) pyramided variety gives longer durability.

The chance of becoming virulence to two or three genes is much less compared to single gene (Mundt *et al.,* 1990). Bacterial blight genes (*Xa1*, *Xa3*, *Xa4*, *xa5*, *Xa7*, *xa13* and *Xa21*) were pyramided into popular varieties, some of them enlisted in Table 1.

Marker assisted selection based released varieties in India

Improved Pusa basmati 1: Pusa Basmati 1 was released in the year 1989 and the variety was popular in cultivation because of high yielding with excellent grain quality. But this variety was highly susceptible to Bacterial Blight disease. Bacterial resistant gene (*xa13* and *Xa21*) were pyramided into Pusa Basmati 1 through Marker Assisted Backcrossing Selection

Table 1. Genes conferring resistance to bacterial blight transferred into popular cultivars of rice using molecular markers.

S.N.	Target gene	Molecular Markers	Crosses	Reference
1	*xa5*, *xa13* and *Xa21*	RG556, RG136, pTA248	PR106 * X IRBB62	Singh *et al*., 2001
2	*Xa21* and *Xa4*	pTA248, STS marker (MP1 and MP2)	MIANHUI * X IRBB24	Ming *et al*., 2006
3	*xa5*, *xa13,Xa21* and *Xa4*	RG556, RG136, pTA248 and STS marker (MP1 and MP2)	Jyothi /IR50* X NH56	Bharathkumar *et al*., 2010
4	*xa13* and *Xa21*	RG136, pTA248	Pusa 6B and PRR78* X PUSA 1460	Basavaraj *et al*., 2010
5	*xa5*, *xa13* and *Xa21*	RG556, RG136, pTA248	ADT43 AND ADT47* X IRBB60	Bharani *et al*., 2010
6	*xa13* and *Xa21*	RG136, pTA248	Type 3 Basmati* x PR 106-P2	Rajpurohit *et al*., 2011
7	*xa13,Xa21,wx* and *sd1*	RG136, pTA248, RM190, RM339 and RM284	Basmati370 and Basmati 386 x IET17948	Bhatia *et al*., 2011
8	*xa13, Xa21, Pi54 and qSBR 11-1*	RG136, pTA248, RM206 and RM224	Improved Pusa Basmati 1* x Tetep	Singh *et al*., 2012
9	*xa5,xa13,Xa21* and *fgr*	RG556, RG136, pTA248 and RM515	IRS5441-2xIRBB59	Salgotra *et al*., 2012
10	*xa13* and *Xa21*	RG136, pTA248	Taraori Basmati and Basmati 386* x Improved Samba Mahsuri (ISM)	Pandey *et al*., 2013
11	*xa13 and Xa21,*	RG136, pTA248	MTU1010* x B95-1 (Improved Samba Mahsuri)	Magar *et al*., 2014
12	*xa5, xa13, Xa21*	RG556, RG136, pTA248	Jalmagna*x Swarna BB pyramid line	Pradhan *et al*., 2015

*Popular cultivar used as recurrent parent to improve resistant to bacterial blight disease.

(Joseph *et al*., 2004). During each backcross generation, plants which had bacterial resistant gene in the background of Pusa Basmati 1 was selected and a series of field trials were conducted. An improved version retains unique characteristics of basmati with target resistant genes (*xa13* and *Xa21*) and released for cultivation in the year 2007 (ICAR, 2009).

Improved samba mahsuri: Samba Mahsuri was popular cultivated in southern states of Indian country, but the variety was susceptible to Bacterial Blight disease. To increase resistance to this disease, Sundaram *et al*. (2008) pyramided three bacterial blight resistant genes (*xa5*, *xa13* and *Xa21*) with the help of gene linked markers (RG556, RG136 and pTA248) into Samba Mahsuri. Improved version possessed disease resistant gene without any comprise in the unique features of the variety and released for commercial cultivation in 2009 (Smart Indian Agriculture, 2016).

Swarna *Sub1*: Swarna was released in 1982 even though the variety has good yielding capacity and it does not withstand flash floods and typhoons. Scientists were introgressed *Sub1A* into Swarna variety by Marker Assisted Backcrossing Strategy. Improved version shows tolerant to complete submergence of about two weeks. Swarna *Sub1* was released in 2009 for cultivation especially low land areas (Bailey-Serres *et al*., 2010)

Suggestive measures for obstacles in development of resistance against blight disease: Breeders have realized potential and benefit of Marker Assisted Selection by improving the efficiency of selection but it also have some disadvantages. The problem of widely used SSR marker is that they require extensive sequence data from the species of interest as a result it leads to increase initial investment in molecular breeding research. But, once marker is validated this should allow more widely applicable for crop breeding programmes (Reddy, 2017). Some of the markers *viz*., RAPD and ISSR have reproducibility problem *i.e.* these types of markers not capable of reproduce result across different laboratories (Kumar *et al*., 2009, Shehata *et al*., 2009). But the amplification products of RAPD are found to be reproducible when the reactions are repeated using the same reaction conditions (Kumari and Thakur, 2014). In field crops, most of economic importance characters like yield and its parameters are complex inheritance because it is governed by several genes. So, it is always a great challenge to develop marker linked to Quantitative Trait Loci (QTL) characters. The performance of QTL is unpredictable because in experimental studies of QTL mapping is restricted only to biparental combination and it may not effective in different back ground because of interaction with loci and epistasis (Holland 2007; Collard *et al*., 2008). The development and accuracy of marker closely linked to QTL depends on the following factor *viz*., population size, extent of genetic variation and number of DNA markers. Resolution can be dramatically improved with several generation intercrossing when establishing MAGIC (Multi-Parent Advanced Generation Intercross: It is defined as interrogate alleles from multi-parent crosses and to provide increased recombination and mapping resolution) or NAM (Nested Association Mapping: It is approach to

the mapping of genes underlying complex traits in which the statistical power of QTL is combined with the high chromosomal resolution of association mapping) (Korte and Farlow, 2013; Bandillo *et al*., 2013; Yu *et al*., 2008).

Cost effectiveness of MAS: MAS has been proven to successful technique for selection of desirable plants in segregation population without linkage drag, pyramiding disease resistance gene and shortens period of releasing new cultivars. But the usage of MAS is restricted to traits with monogenic inheritance, laboratory with highly technical equipment and well trained scientific human resource as well as operation resource (Nilausen *et al*., 2016; Yang *et al*., 2016). The reliability and reproducibility of QTL based marker is unpredictable because of these types of markers were developed only in limited period of time/locations. So, the success of this technique depends on factors that influence markers of heritability trait, phenotypic screening and cost of inputs. Once marker developed and validated for concerned trait it is cheaper than conventional breeding (Roychowdhury *et al*., 2013). In comparing cost effectiveness of conventional and MAS, it is necessary to consider the value of time savings by MAS and accelerated release of varieties turn into economic benefits. So, integrating the knowledge of both molecular biologist and plant breeders will enable the effective application in plant breeding programmes (Mba *et al*., 2012).

Conclusion

After the successful release of Marker Assisted Selection based varieties for cultivation, the significance of molecular marker has become popular all over the world. Molecular markers not improve the efficiency of plant breeding method but also it saves money and time. In future days, breeders have choice of using traditional breeding methodologies in combination with marker assisted selection to release varieties in short period of time.

REFERENCES

Agarwal, P., Parida, S.K., Raghuvanshi, S., Kapoor, S., Khurana,P., Khurana, J. P. and Tyagi, A.K. (2016). Rice improvement through genome-based functional analysis and molecular breeding in India. *Rice* 9:1

Agricultural Statistics at a glance (2014). Retrieved 2015 from https://eands.dacnet.nic.in/agricultural statistics at glance 2014.pdf

Arunakumari, K., Durgarani, C.V., Satturu, V., Sarikonda, K.R., Chitoor, P.D.R., Vutukuri, B., Laha, G.S., Nelli, A.P.K., Gattu, S., Jamal, M., Prasadbabu, A., Hajra, S. and Sundaram, R.M. (2016) Marker-Assisted Pyramiding of Genes Conferring Resistance Against Bacterial Blight and Blast Diseases into Indian Rice Variety MTU1010. *Rice Science* 23: 306-316

Bailey-Serres, J., Fukao, T., Ronald, P., Ismail, A., Heuer, S. and Mackill, D. (2010). Submergence tolerant rice: Sub1 journey from landrace to modern cultivars. *Rice* 3: 138-147.

Bandillo, N., Raghavan, C., Muyco, P.A., Sevilla, M.A.L., Lobina, I.T., Ermita, J.D., Tung, C.W., Mccouch, S., Thomson, M,m Mauleon, R, Singh, R.K., Gregoria, E.R. and Leung, H. (2013). Multi parent Advanced Generation Inter Cross (MAGIC) populations in rice: progress and potential for genetic research and breeding. *Rice* 6:11

Basavaraj, S. H., Singh, V.K., Singh, A., Singh, A., Singh, A., Anand, D., Yadav, S., Ellur, R.K., Singh, D., Krishana, S.G., Nagarajan, M., Mohapatra, T., Prabhu, K. V. and Singh, A.K. (2010). Marker assisted improvement of bacterial blight resistance in parental lines of Pusa RH10 a superfine grain aromatic rice hybrid. *Mol Breed* 26: 293–305.

Bernatzky, R. and Tanksley, S.D. (1986). Methods for detection of single or low copy sequences in tomato on southern blots. *Plant Mol Biol Rep* 4: 37-41.

Bharani, M., Nagarajan, P., Rabindran, R., Saraswathi, R., Balasubramanian, P. and Ramalingam, J. (2010). Bacterial leaf blight (*Xa21*, *xa13* and *xa5*) pyramiding through molecular marker assisted selection into rice cultivars. *Arch of Phytopathology and Plant Protection* 43: 1032-1043

Bharathkumar, S., Paulraj, R. D., Brindha, P. V., Kavitha, S. and Gnanamanickam, S.S. (2010). Improvement of bacterial blight resistance in rice cultivars Jyothi and IR50 via Marker-Assisted Backcross Breeding. *J Crop Improv* 21: 101–116.

Bhatia, D., Sharma, R., Vikal, Y., Mangat, G.S., Mahajan, R., Sharma, N., Lore, J.S., Singh, N., Bharaj, T.S. and Singh, K. (2011). Marker-Assisted development of Bacterial Blight resistant, dwarf and high yielding versions of two traditional basmati rice cultivars. *Crop Sci* 51: 759-770.

Causse, M.A., Fulton, T.M., Cho, Y.G., Ahn, S.N., Chunwongse, J., Wu, K.S., Xiao, J.H., Yu, Z.H., Ronald, P.C., Harrington, S.E. (1994). Saturated molecular map of the rice genome based on an interspecific backcross population. *Genetics*. 138:1251–1274.

Charpe, A., Koul, S., Gupta, S. K., Singh, A., Pallavi, J.K. and Prabhu K. V. (2012). Marker assisted gene pyramiding of leaf rust resistance genes *Lr9*, *Lr24* and *Lr28* in a bread wheat cultivar HD 2329. *J. Wheat Res.* 4 20–28.

Crop Improvement (2009). DARE/ICAR annual report, Indian Council of Agricultural Research (ICAR), New Delhi pp.29-40.

Collard, B.C.Y. and Mackill, D.J. (2008) Marker assisted selection: an approach for precision plant breeding in twety-first century. *Philos Trans R Soc Lond B Biol Sci*. 363: 557–572

Cuc, L.M., Huyen, L.T.N., Hien, P.T.M., Hang, V.T.T., Dam, N.Q., Mui, P.T., Quang, V.D., Ismail, A.M. and Ham, L. H. (2015) 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* 3:528-536

Holland, J.B. (2007) Genetic architecture of complex traits in plants. *Current Opinion in Plant Biology*. 10:156–161.

Huq, M.A.,Akter, S., Nou, S., Kim, H.T., Jung, Y.J. and Kang, K.K. (2016) Identification of functional SNP in genes andtheir effect on plant phenotype. *J of Plt Bio-*

tech. 43: 1-11.

Joseph, M., Gopalakrishnan, S., Sharma, R.K., Singh, V.P., Singh, A.K., Singh, N.K. and Mohapatra, T. (2004). Combining Bacterial Blight resistance and Basmati quality characteristics by phenotypic and molecular Marker-Assisted Selection in rice. *Mol Breed* 13: 377–387.

Korte, A. and Farlow, A. (2013). The advantage and limitations of trait analysis with GWAS: a review. *Plant Methods* 9: 29

Kumar, P., Gupta, V.K., Misra, A.K., Modi, D.R. and Pande, B.K. (2009). Potential of Molecular Markers in Plant Biotechnology. *Plant Omics Journal* 2:141-162.

Kumari, N. and Thakur, S. K. (2014). Randomly Amplified Polymorphic DNA a brief review. *American J of Animan and Vetrinary Science* 9(1): 6-13

Kurokawa, Y., Noda, T., Yomagata, Y., Angeles-shim, R., Sunohara, H., Vehara, K., Furuta, T., Nagai, K. and Kazuyuki, D. (2016). Construction of a versatile SNP array for pyramiding useful gene of rice. *Plant Science* 242: 131-139.

Lateef, D.D. (2015). DNA marker technologies in plants and applications for crop improvements. *Journal of Bioscience and Medicines* 3: 7-18

Magar, M.M., Rani, D. and Anuradha, G. (2014). Marker Assisted Selection for Bacterial leaf Blight resistance in segregating populations of *Cottondora sannalu*. *Int J Appl Sci Biotechnol*, 2:229-237.

Mammadov, J., Aggarwal, R., Buyyarapu, R. and Kumpatla, S. (2012) SNP markers and their impact on plant breeding. *Int J of Plant Genomics*. 2012: 1-11.

Mba, C., Guimaraes, E, P and Ghosh, K. (2012). Re-orienting crop improvement for the changing climatic condition of the 21st century. *Agri and Food Security*. 1:7

Mehrotra, R. S.and Agarwal, A. (2003). Plant Pathology. Tata McGraw-Hill, New Delhi.

McCouch, S.R., Kochert, G., Yu, Z.H., Wang, Z.Y., Khush, G.S., Coffman, W.R. and Tanksley, S.D. (1988) Molecular mapping of rice chromosomes. *Theor Appl Genet.* 76:815–829.

Ming, D.Q., Quan, W.S., Ping, Z.A., Yu, Z.H. and Ping, L. (2006). Breeding rice restorer lines with high resistance to Bacterial Blight by using molecular Marker-Assisted Selection. *Rice Sci* 13: 22–28.

Mundt, C.C. (1990). Probability of mutation to multiple virulence and durability of resistance gene pyramids. *Phytopathol* 80: 221-223

Nilausen, C., Gelinas, N. and Bull, G. (2016). Perceived acceptability of implementing marker assisted selection in the forest of British Colubia. *Forest* 7:286

Orjuela, J., Garavito, A., Bouniol, M., Arbelaez, J.D., Moreno, L., Kimball, J., Wilson, G., Rami, J.F., Tohme, J., Mccouch, S.R. and Lorieux. M. (2010). A Universal Core Genetic Map of rice. *Theor Appl Genet* 120: 563-572.

Oste, C. (1989) PCR technology. Principles and applications for DNA amplification. Palgrave Macmillam UK. Pp. 23-30.

Pandey, M.K., Rani, N.S., Sundaram, R.M., Laaha, G.S., Madhav, M.S., Rao, K.S., Sudharshan, I., Hari, Y., Varaprasad, G.S., Rao, L.V.S., Suneetha, K., Sivaranjani, A.K.P., Viraktamath, B.C. (2013). Improvement of two traditional Basmati rice varieties for bacterial blight resistance and plant stature through morphological and marker-assisted selection. *Mol Breeding* 31:239–246.

Pradhan, S.K., Nayak, D.K., Mohanty, S., Behera, L., Barik, S.R., Pandit, E., Lenka, S. and Anandan, A. (2015). Pyramiding of three Bacterial Blight resistance genes for broad-spectrum resistance in deepwater rice variety, Jalmagna. *Rice* 8:19

Rajpurohit, D., Kumar, R., Kumar, M., Paul, P., Awasthi, A., Basha, P.O., Puri, A., Jhang, T., Singh, K. and Dhaliwal, H.S. (2011). Pyramiding of two Bacterial Blight resistance and a semi dwarfing gene in Type 3 Basmati using Marker-Assisted Selection. *Euphytica* 178: 111-126.

Reddy, V.R.P. (2017). New concepts in plant breeding and genetics. *Adv in Plants and Agri Res* 7(1): 00245.

Roychowdhury, R., Taoutou, A., Harseem, K. R. and Tan, J. (2013) Molecular marker assisted technologies for crop improvement. Crop Improvement in the era of climate change. International Publication House Ltd New Delhi 241-258.

Saiki, R.K., Gelfand, D.H., Stoffel, S., Scharf, S.J., Higuchi, R., Horn, G.T., Mullis, K.B. and Erlich, H.A. (1988) Primer-directed enzymatic amplification of DNA with a thermostable DNA polymerase. *Science* 239: 487-491.

Salgotra, R.K., Gupta, B.B., Millwood, R.J., Balasubramaniam, M., Stewart, C.N. (2012). Introgression of Bacterial Leaf Blight resistance and aroma genes using functional Marker-Assisted Selection in rice (*Oryza sativa* L.). *Euphytica* 187:313–32.

Sanchez, A. C., Brar, D. S., Huang, N. and Khush, G.S. (2000). Sequence Tagged Site markers-assisted selection for three Bacterial Blight resistance genes in rice. *Crop Sci* 40: 792–797

Shanthi, M.L., Devi, G.L., Kumar, G.N. and Shashidhar, H.E. (2010). Molecular Marker-assisted selection: a tool for insulating parental lines of hybrid rice against Bacterial leaf Blight. *Intl J Plant Pathology* 1: 114–123.

Shehata S.M., Ammar, M.H., Abdelkalik, A.F. and Zayed, B.A. (2009). Morphological, molecular and biochemical evaluation of Egyptian jasmine rice variety and its M5 derived mutants. *Afr J Biotechnol* 8: 6110-6116

Singh, S., Sidhu, J.S., Huang, N., Vikal, Y., Li, Z., Brar, D.S , Dhaliwal, H.S. and Khush, G.S. (2001). Pyramiding three Bacterial Blight resistance genes (*xa5*, *xa13* and *Xa21*) using Marker-Assisted Selection into indica rice cultivar PR106. *Theor Appl Genet* 102: 1011 – 1015.

Singh, A. and Sengar, R.S. (2015). DNA Fingerprinting Based Decoding of Indica Rice (*Oryza sativa* L) *via* Molecular Marker (SSR, ISSR, & RAPD) in Aerobic Condition. *Adv Crop Sci* Tech 3:2.

Smart Indian Agriculture (2016) In Conversation with Ramesh Sonti, Lead Developer of Improved Samba Mashuri Rice With Bacterial Blight Resistance http://www.smartindianagriculture.in/

Sundaram, R.M., Vishnupriya, M.R., Biradar, S.K., Laha, G.S., Reddy, G.A., Rani, N.S., Sarma, N.P. and Sonti, R.V. (2008). Marker assisted introgression of bacterial blight resistance in Samba Mahsuri, an elite indica rice variety. *Euphytica* 160: 411–422.

Temnykh, S., Decterck, G., Lukashova, A., Lipovich, L., Cartinhour, S. and Mccouch, S. (2001). Computational and experimental analysis of microsatellites in rice

(*Oryza sativa* L.): Frequency, length variation, transposon associations and genetic marker potential. *Genome Res* 11: 1441-1452.

Toledo, A.M., Ignacio, J.C., Casal, C., Gonzaga, Z.J., Mendioro, M. and Septiningsih, E. (2015) Development of improved Ciheang-Sub1 having tolerance to anaerobic germination conditions. *Plant Breed Bio* 3: 77-87

Vos, P., Hogers, R., Bleeker, M., Reijans, M., Lee, V., Hornes, M., Frijters, A., Pot, J., Peleman, J. and Kuiper, M. (1995). AFLP: a new technique for DNA fingerprinting. *Nucleic Acids Res* 23: 4407-4414.

Wijerathna, Y.M.A.M. (2015) Marker Assisted Selection: Biotechnology for rice molecular breeding. *Adv in Crop Science and Technology*. 3(4): 1000187

Williams, J.G.K., Kubelik, A.R., Livak, K.J., Rafalski, J.A. and Tingey, S.V. (1990). DNA polymorphisms amplified by arbitrary primers are useful as genetic markers. *Nucleic Acids Res* 18: 6531-6535.

World population prospects the 2015 Revision. Retrived 2015 from https://esa.un.org/unpd/wpp/publications/files/key_findings_wpp_2015.pdf

Yang, S., Fresnedo-Ramrez, J., Wang, M., Cote, L., Schweitzer, P., Barba, B., Takacs, E.M., Clark, M., Lyby, J., Manns, D.C., Sacks, G., Mansheld, A.K., Londo, J., Fennelle, A., Gadoury, D., Reisch, B., Dvidson, L.C. and Sun, Q. (2016). A next-generation marker genotyping platform (Ampseq) in heterozygous crops: a case study for MAS in grape vine. *Hort Res.,* 3:16002

Yu, J., Rolland, J.B., Mcmullen, M.D. and Bucker, E.S. (2008). Genetic design and statistical power of Nested Association mapping in maize. *Genetics* 178: 539-551.

Zhang, G., Angeles, E.R., Abenes, M.L.P., Khush, G.S. and Huang, N. (1996). RAPD and RFLP mapping of the bacterial blight resistance gene *xa13* in rice. *Theor Appl Genet* 93:65–70