Assessment of parental polymorphism between Swarna / *Oryza rufipogon* IC309814 derived mapping population

Abstract

Detection of molecular markers with parental polymorphism is a pre-requisite for QTL mapping and further identification of genomic regions. PCR based InDel markers tend to have a greater degree to show polymorphism among the parental genotypes for any specific cross. Polymorphic studies were carried out between the two parents Swarna (cultivated rice) and *Oryza rufipogon* IC309814 (Wild rice) using 192 randomly selected InDel markers and 40 gene specific markers for Iron and Zinc covering the entire 12 chromosomes. Results revealed that InDel markers were polymorphic between both the parents and highest percentage of polymorphism observed on chromosome 10 (75%) followed by chromosome 5 (60%) and chromosome 9 (55.6%) and lowest polymorphism percentage was shown in chromosome 3 (8.3%) with an overall 22.4 % polymorphism percentage among all the markers used in this study. Detected polymorphic (SSR) markers were for genotyping the mapping population derived from the cross between Swarna cultivar and *Oryza rufipogon* to map Quantitative Trait Loci (QTLs) associated with yield and grain quality traits.

Keywords: Swarna, Wild Rice, Parental Polymorphism, InDels and Biofortification

UNDER PEER REVIEW

Introduction

Rice, botanically called as *Oryza sativa* L. and belongs to poaceace family, it serves as a primary food source as well as staple cereal crop for population globally. The crop is extensively cultivated in the Asian countries having major share in terms of production (Fukagawa *et al.*, 2019). Some wild relatives of rice found to have higher grain Fe and Zn concentrations compared with the cultivated rice germplasm (Garcia- Oliveira *et al*., 2018). The AA-genome, wild progenitor of cultivated rice *O. rufipogon* is a rich source of natural allelic variation for several agronomic, grain quality and grain micronutrient trait. Wild accessions *O. nivara* and *O. rufipogon* have high concentrations of grain iron and zinc (Anuradha *et al*., 2012a)

Genetic markers have helped in plant breeding methodologies by enabling precise tracking of trait inheritance and providing robust tools for genetic diversity assessment (Chakravarthi *et al*.,2006). These molecular markers are environment-independent in nature and helps to

understand genetic similarities and characterization. When such markers are very tightly linked to genes of interest, they can be used for the indirect selection of desirable alleles, which is known as marker-assisted selection (Yu *et al.,* 2005).

Parental polymorphism is a prerequisite for implementing marker-assisted selection (MAS) and marker-assisted backcross breeding (MABB) methodologies. Molecular genetic analyses are essential to establish allelic diversity and detect polymorphic loci that facilitate introgression of quantitative trait loci (QTLs) from donor germplasm into recipient genetic backgrounds. Without demonstrable polymorphism across target genomic regions, the identification and subsequent selection of progenies harboring specific traits become fundamentally unachievable (Yerva *et al.,*2018). The genetic variation assessment provides the foundational framework for successful breeding strategies, ensuring the potential for trait introgression and genetic improvement in crop improvement programs (Graham *et al*., 1999).

Polymorphism levels depends on the genetic diversity of genotypes and primer specificity during DNA sequence recognition. .These polymorphic markers are pivotal for genetic diversity assessment, linkage map construction, gene mapping, and marker-assisted selection (MAS) (Edwards and Batley, 2010; Gonzaga *et al.,* 2015).

In marker-assisted breeding, InDel markers offer advantages over SSRs and SNPs. These markers generate fewer shadow bands due to their conservative polymorphic regions, enabling more precise genotype characterization. The enhanced reliability and specificity of InDel markers make them a valuable tool for genetic mapping and breeding strategies (Mo *et al*., 2024). QTL analysis allows detection of marker-trait associations, revealing genetic control of complex traits. By utilizing markers linked to QTLs, rice breeders can implement precise selection strategies, positively selecting beneficial elements and negatively selecting potentially undesirable components (Rathod *et al.,* 2021). QTL mapping enables precise dissection of complex genetic regions, facilitating targeted breeding strategies (Swamy *et al.,* 2018).

The objective of this study is to identify polymorphic InDel markers showing genetic variation between the cultivated rice variety (Swarna) and the wild rice accession (*Oryza rufipogon* IC309814). These polymorphic markers would be crucial for mapping quantitative trait loci (QTLs) in the advanced backcross population derived and further to identify the genomic regions associated with specific traits.

Materials and methods

UNDER PEER REVIEW

The experimental material constitutes of two rice genotypes i.e., most popular mega cultivar Swarna and accession IC 390814 of wild species *Oryza rufipogon* available at Indian Institute of Rice Research (IIRR). From both the genotypes genomic DNA was isolated using CTAB method (Doyle and Doyle, 1987). In the procedure, 0.1g of young and tender leaves were used for grinding using mortar and pestle (Maxprep) with CTAB buffer (4% CTAB, 100 mM Tris HCl, 20 mM EDTA, 1.4 M NaCl). The DNA concentration is quantified at OD values of 260 and 280 nm using nanodrop to check the purity and quantity of genomic DNA.

In all, 192 randomly selected InDel markers covering all the 12 chromosome and 40 gene specific markers (Table no.) linked to Iron and Zinc concentration were used in the polymorphic studies between Parent 1 (Swarna) and Parent 2 (*O. rufipogon* IC 309814). The primer sequence and chromosomal location information were retrieved from Hechanova *et al*., 2022.

PCR reaction was carried out in thermal cycler (Biorad Thermal cycler) with a final reaction volume of 10µl containing 30ng of genomic DNA, Master mix, 10 pmol of forward and reverse primers. The programme followed for PCR products as: initial denaturation at 94°C for 7 min followed by 35 cycles of 94°C for 30 s, 55°C (58°C for gene-specific markers) for 30 s, 72°C for 1 min, and a final extension of 10 min at 72°C. Amplified products were resolved in 2.5 % agarose gel prepared in 1X TAE buffer and electrophoresed at 150 V for 1 hour. Gels were stained with ethidium bromide and documented using a gel documentation system (Vilber- BIO-PRINT cx4 Edge- Fixed pad Container, France).

The percentage of polymorphism calculated using the formula:

$$\text{Polymorphism \%} = \frac{\text{RM Primers with Polymorphism}}{\text{Total number of RM primers used}} \times 100$$

Table 1. Characteristics of Parental lines used in this study

Feature	**Swarna (Cultivated)**	***Oryza rufipogon* IC309814 (Wild)**
Origin	India	Native to Southeast Asia

UNDER PEER REVIEW

Genetic Background	Cultivated species (*Oryza sativa*)	Wild species (*Oryza rufipogon*)
Morphology	Semi-dwarf, robust plants	Typically shorter to tall, variable in size
Plant Height	Typically 90-120 cm	Generally shorter, around 60-100 cm
Leaf Color	Dark Green, broader leaves	Narrower, often darker green
Growth Habit	Semi-dwarf, erect	More variable, can be erect or prostrate
Grain Color	White to pale yellow	Dark brown to reddish-brown
Grain Shape	Long and slender	Shorter, often irregular
Grain Length	6.5-7.5 mm	5-6 mm
Grain Breadth	2.0-2.5 mm	1.5-2.0 mm
Kernel Length	6.0-7.0 mm	5.0-6.0 mm
Kernel Breadth	2.0-2.4 mm	1.4-1.8 mm
L/B Ratio	2.8-3.0	3.0-4.0
Disease Resistance	Moderate	High, evolved resistance to major biotic stresses
Yield Potential	High, suited for cultivation	Low, variable in wild habitats
Adaptation	Prefers flooded conditions	Adapted to diverse environments
Genetic Diversity	Pure line developed through selective breeding	High, significant genetic variability
Conservation Status	Widely cultivated, not endangered	Often threatened due to habitat loss

Source: Swarna (Haug *et al.,* 2004, IRRI 2017), *Oryza rufipogon* (Wang *et al.,* 2008, Vaughan *et al.,* 1997)

Parent 1 Swarna **Parent 2 *Oryza rufipogon* IC 390814**

UNDER PEER REVIEW

Fig 1:Difference in the plant and grain phenotype of Parent1 (Swarna) and Parent 2 (*Oryza rufipogon* IC 390814)

Results and Discussion

The quantity of isolated DNA samples checked using Nanodrop from both the parents its concentration ranged between 2108 to 2765 ng/ µl, were it is further diluted in a ratio of 1:20 for PCR reaction. Ratio of UV absorbance of DNA at OD 260/280 ranges from 1.80 to 1.97 and hence the DNA samples are standard and good. Using the diluted genomic DNA of the both parents, we have screened 192 InDel markers and 40 gene specific markers for Fe and Zn concentration across all the chromosomes, 52 InDel markers were polymorphic between Swarna and *O rufipogon* IC 309814 with 22.4 % of polymorphism percentage. None of the gene specific markers are polymorphic among the parents. The polymorphic markers along with their chromosomes depicted in **Table 1 and Fig 2**. Similar type of studies were done previously by Shankar *et al.,* (2023) and identified 85 polymorphic markers between parents Jyoti and Chuvannnamaran. In another study focused on iron and zinc content, *Shivani et al.,* (2020) reported 52 polymorphic SSRs among the 171 markers used for iron and zinc content between the Swarna and type 3, Bimpong *et al.,* 2011 reported 40.5 percent of polymorphism among the *O. sativa* cv. IR64 and *O. glaberrima* parents using 464 markers. Above findings reveals that the varying degrees of genetic diversity that can be detected between different rice varieties using SSR markers, with polymorphism rates ranging from approximately 30-41% across these different parental combinations.

Swamy *et al.,* 2018 have reported100 polymorphic markers between Swarna /*O.nivara* (IRGC81832), Hu et al 2014 also reported 108 polymorphic markers between Xieqingzao B (*O.sativa*) / *O.rufipogon*. 101 polymorphic markers were identified by Anuradha *et al.,* 2012b between Madhukar/ Swarna. Ilango and Sarla, (2010) have studied parental polymorphism between 5 parental lines Swarna, BPT5204, Madhukar, Jalmagna, and IR64 using 112 RM markers out of which 33 polymorphic markers were reported.

UNDER PEER REVIEW

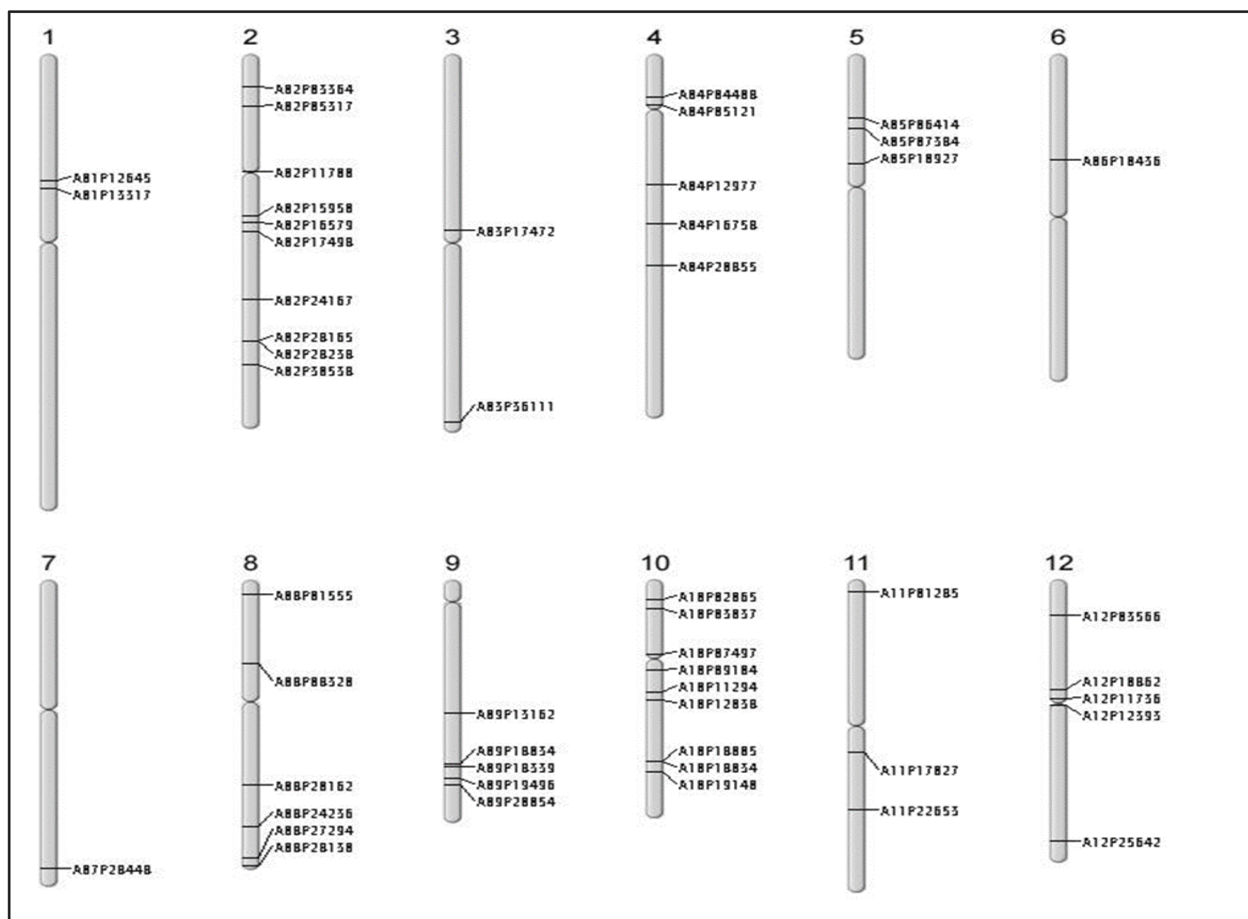

Fig 2. Map image showing physical location and distribution of 52 polymorphic markers

Chromosome wise polymorphism percentage of InDel markers is showed in the **Table 2.** Among the 192 markers Chromosome 10 had the highest polymorphism percentage i.e., 75% (9 markers- A10P02065, A10P03037, A10P07497, A10P09104, A10P1129, A10P12038, A10P18005, A10P18034, A10P1910) followed by chromosome 4 with 60% (5 markers-A04P04408, A04P05121, A04P12977, A04P16758, A04P20855), Chromosome 9 with 55.6% (5 markers- A09P1312, A09P1803, A09P18339, A09P19496, A09P20054) and lowest percentage of polymorphism was observed in chromosome 3 with 8.3 % (2 markers- A03P17472, A03P36111). Frequency distribution of polymorphic markers are shown in **Fig no. 3**. Polymorphic studies conducted by Yerva *et al.,* 2108., Shankar *et al.,* 2020, and Singh *et al.,* 2022 in rice consistently demonstrated that molecular markers exhibit varying frequencies across different rice chromosomes which are reported in their studies.

Table 2. List of Polymorphic markers between the Swarna and *O rufipogon* IC 309814

S.No.	Marker	Chr No.	Amplicon Size	S.No.	Marker	Chr No.	Amplicon Size

1	**A01P12645**	1	173
2	**A01P13317**	1	138
3	**A02P03364**	2	204
4	**A02P05317**	2	164
5	**A02P11700**	2	132
6	**A02P15950**	2	133
7	**A02P16579**	2	276
8	**A02P17498**	2	288
9	**A02P24167**	2	279
10	**A02P28165**	2	140
11	**A02P28238**	2	261
12	**A02P30538**	2	170
13	**A03P17472**	3	406
14	**A03P36111**	3	187
15	**A04P04408**	4	366
16	**A04P05121**	4	256
17	**A04P12977**	4	274
18	**A04P16758**	4	656
19	**A04P20855**	4	220
20	**A05P06414**	5	276
21	**A05P07384**	5	352
22	**A05P10927**	5	208
23	**A06P10436**	6	281
24	**A07P28448**	7	127
25	**A08P01555**	8	160
26	**A08P08320**	8	131
27	**A08P20162**	8	213
28	**A08P24236**	8	177
29	**A08P27294**	8	376
30	**A08P28130**	8	118
31	**A09P13162**	9	134
32	**A09P18034**	9	219
33	**A09P18339**	9	309
34	**A09P19496**	9	224
35	**A09P20054**	9	146
36	**A10P02065**	10	189
37	**A10P03037**	10	163
38	**A10P07497**	10	164
39	**A10P09104**	10	147
40	**A10P11294**	10	161
41	**A10P12038**	10	255
42	**A10P18005**	10	173
43	**A10P18034**	10	180
44	**A10P19140**	10	179
45	**A11P01285**	11	213
46	**A11P17027**	11	237
47	**A11P22653**	11	291
48	**A12P03566**	12	230
49	**A12P10862**	12	212
50	**A12P11736**	12	222
51	**A12P12393**	12	193
52	**A12P25642**	12	334

Table 3 Chromosome wise polymorphism percentage of RM markers

Chromosome no.	Total No. of SSR marker used	No. of Polymorphic markers obtained	Polymorphism (%)
Chromosome 1	14	2	14.3
Chromosome 2	25	10	40.0
Chromosome 3	**24**	**2**	**8.3**
Chromosome 4	20	5	25.0
Chromosome 5	5	3	60.0
Chromosome 6	8	1	12.5
Chromosome 7	7	1	14.3
Chromosome 8	23	6	26.1
Chromosome 9	9	5	55.6
Chromosome 10	**12**	**9**	**75.0**
Chromosome 11	19	3	15.8
Chromosome 12	26	5	19.2
Total	**192**	**52**	**22.4**

UNDER PEER REVIEW

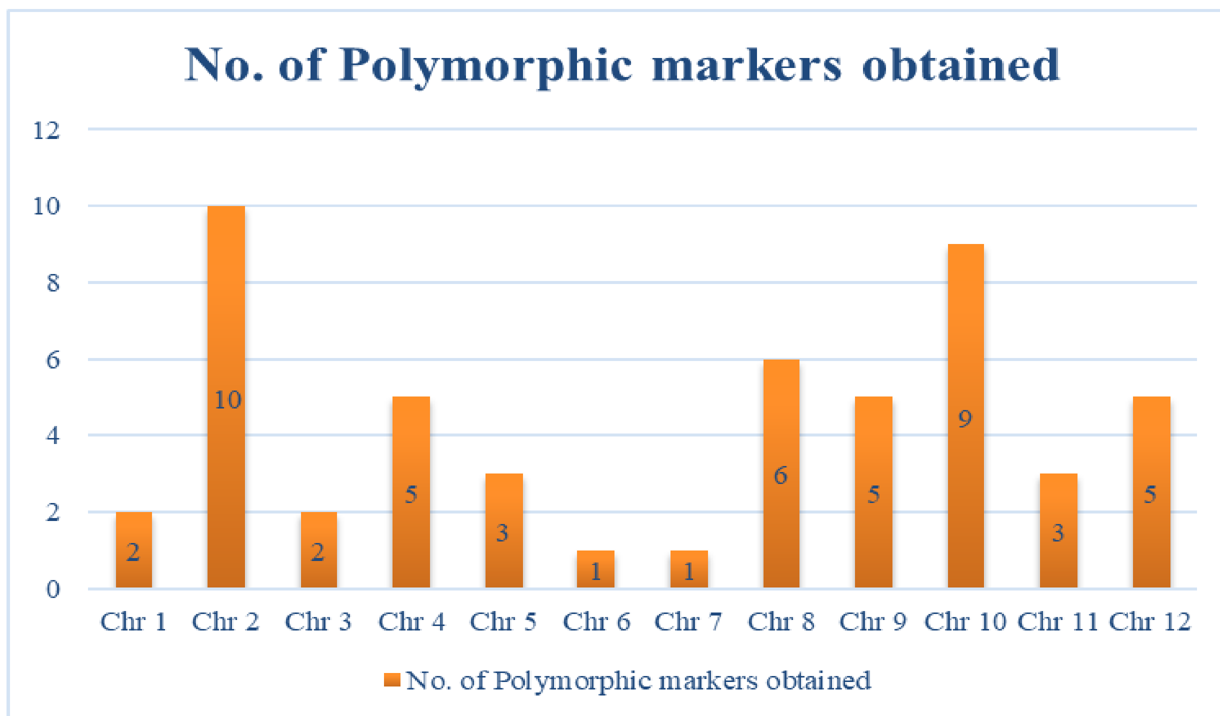

Fig 3. Frequency distribution of polymorphic markers across the chromosomes

Amplicon size of markers ranged from 117 bp to 656 bp, and polymorphic markers were 52 out of 232 and the remaining were monomorphic as showed in **Fig 4 and 5**. Similar type of banding pattern were reported by Chandu *et al.,* 2020, Rathi *et al.,* 2021 , Majhi *et al.,* 2022 and Shankar *et al.,* 2023 for the markers used in this study.

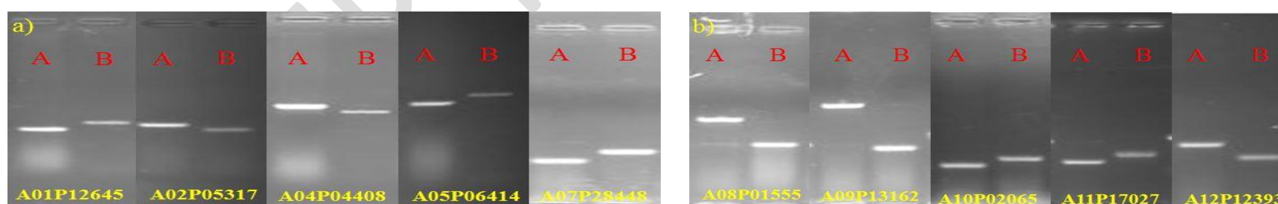

Fig 4. Polymorphic markers between the parents

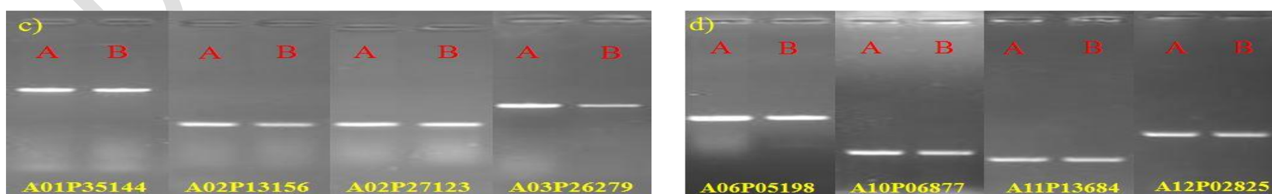

Fig 5. Monomorphic markers between the parents

Genotypic data using polymorphic markers are useful for linkage studies and diversity analysis for a number of linked characteristics in rice using the detected polymorphic molecular markers. Polygenic traits, which were previously difficult to analyze, using traditional breeding methods, would now be easily tagged using molecular markers. The screening of markers for parental polymorphism among the rice cultivars forms the basis for tagging of the desired gene, fine mapping of the gene in the rice chromosome and in the subsequent Marker Assisted Selection (MAS) programmes. The characterization and physical mapping of polymorphic InDel markers represents a significant advancement in molecular breeding, by establishing a well-defined set of these markers, which serve as genotyping of mapping populations to the identification and analysis of quantitative trait loci. Additionally, their application in marker-assisted selection significantly fasten the breeding process, allowing for more precise and efficient selection of desired traits (Hasan *et al.,* 2021, Malabanan-Bauan *et al.,* 2023).

It ultimately contributing to the acceleration of rice variety development and crop improvement programs. The polymorphic rice markers can be further used in fine mapping for iron and zinc rich micronutrient genes in the mapping population obtained from these parents (Welch and Graham, 2004). The present work gives the preliminary understanding the genetic polymorphism exist in the population for biofortification of micronutrient such as iron and zinc in cereals like rice.

Conclusion

This study detected 52 polymorphic markers out of 192 InDel and 40 gene specific markers for iron and zinc were found with a polymorphic percentage of 27.1%, these polymorphic markers can be used in QTL mapping for yield and grain quality traits. Additional polymorphic studies and detection of genome wide polymorphic markers uniformly covering all chromosomes are required to identify precise detection of effective QTLs for target traits.

UNDER PEER REVIEW

Disclaimer (Artificial intelligence)

Option 1:

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc.) and text-to-image generators have been used during the writing or editing of this manuscript.

Option 2:

Author(s) hereby declare that generative AI technologies such as Large Language Models, etc. have been used during the writing or editing of manuscripts. This explanation will include the name, version, model, and source of the generative AI technology and as well as all input prompts provided to the generative AI technology

Details of the AI usage are given below:

1.

2.

3.

References

Anuradha, K., Agarwal, S., Batchu, A.K., Babu, A.P., Swamy, B.M., Longvah, T. and Sarla, N., 2012b. Evaluating rice germplasm for iron and zinc concentration in brown rice and seed dimensions. *J. phytol*, *4*(1), pp.19-25.

Anuradha, K., Agarwal, S., Rao, Y.V., Rao, K.V., Viraktamath, B.C. and Sarla, N., 2012a. Mapping QTLs and candidate genes for iron and zinc concentrations in unpolished rice of Madhukar× Swarna RILs. *Gene*, *508*(2), pp.233-240.

Bimpong, I.K., Serraj, R., Chin, J.H., Ramos, J., Mendoza, E.M., Hernandez, J.E., Mendioro, M.S. and Brar, D.S., 2011. Identification of QTLs for drought-related traits in alien introgression lines derived from crosses of rice (*Oryza sativa* cv. IR64)× *O. glaberrima* under lowland moisture stress. *Journal of Plant biology*, *54*, pp.237-250.

Chakravarthi, B.K. and Naravaneni, R., 2006. SSR marker based DNA fingerprinting and diversity study in rice (*Oryza sativa*. L). *African Journal of Biotechnology*, *5*(9).

Chandu, G., Addanki, K.R., Balakrishnan, D., Mangrauthia, S.K., Sudhakar, P., Satya, A.K. and Neelamraju, S., 2020. SSR markers for grain iron zinc and yield-related traits polymorphic between Samba Mahsuri (BPT5204) and a wild rice *Oryza rufipogon*. *Electronic Journal of Plant Breeding*, *11*(03), pp.841-847.

Doyle, J.J. and Doyle, J.L., 1987. A rapid DNA isolation procedure for small quantities of fresh leaf tissue. *Phytochemical bulletin*.

Edwards, D. and Batley, J., 2010. Plant genome sequencing: applications for crop improvement. *Plant biotechnology journal*, *8*(1), pp.2-9.

UNDER PEER REVIEW

Fukagawa, N.K. and Ziska, L.H., 2019. Rice: Importance for global nutrition. *Journal of nutritional science and vitaminology*, *65*(Supplement), pp.S2-S3.

Garcia-Oliveira, A.L., Chander, S., Ortiz, R., Menkir, A. and Gedil, M., 2018. Genetic basis and breeding perspectives of grain iron and zinc enrichment in cereals. *Frontiers in Plant Science*, *9*, p.937.

Gonzaga, Z.J., Aslam, K., Septiningsih, E.M. and Collard, B.C., 2015. Evaluation of SSR and SNP markers for molecular breeding in rice.

Graham, R., Senadhira, D., Beebe, S., Iglesias, C. and Monasterio, I., 1999. Breeding for micronutrient density in edible portions of staple food crops: conventional approaches. *Field crops research*, *60*(1-2), pp.57-80.

Hasan, N., Choudhary, S., Naaz, N., Sharma, N. and Laskar, R.A., 2021. Recent advancements in molecular marker-assisted selection and applications in plant breeding programmes. *Journal of Genetic Engineering and Biotechnology*, *19*(1), p.128.

Haug, W. & Subramanian, K., 2004. Rice Varieties in India: A Comprehensive Study. Journal of Plant Breeding, 12(3), pp.45-58.

Hechanova, S.L., Bhattarai, K., Simon, E.V., Clave, G., Karunarathne, P., Ahn, E.K., Li, C.P., Lee, J.S., Kohli, A., Hamilton, N.R.S. and Hernandez, J.E., 2021. Development of a genome-wide InDel marker set for allele discrimination between rice (*Oryza sativa*) and the other seven AA-genome Oryza species. *Scientific reports*, *11*(1), p.8962.

Hu, B.L., Huang, D.R., Xiao, Y.Q., Fan, Y.Y., Chen, D.Z. and Zhuang, J.Y., 2016. Mapping QTLs for mineral element contents in brown and milled rice using an *Oryza sativa*× *O. rufipogon* backcross inbred line population. *Cereal Research Communications*, *44*(1), pp.57-68.

Ilango, S. and Sarla, N., 2010. Microsatellite marker polymorphism in rice varieties rich in iron and zinc endosperm. *Asian Journal of Experimental Biological Science*, *1*(4), pp.751-757.

International Rice Research Institute (IRRI). (2017). "Swarna - Variety Profile." Retrieved from IRRI

Majhi, P.K., Singh, S.K., Anandan, A., Khaire, A.R., Korada, M., Habde, S.V., Singh, D.K., Bhagvan, A.P., Jayasudha, S. and Singh, A., 2022. Parental polymorphism survey for

UNDER PEER REVIEW

evaluation and selection of contrasting parents for drought tolerance in rice (*Oryza sativa* L.) by using SSR markers. *Int J Environ Clim*, *12*(11), pp.2507-19.

Malabanan-Bauan, K.B., Hechanova, S.L., Ahn, E.K., Li, C.P., Choi, I.R., Hernandez, J.E., Jena, K.K. and Kim, S.R., 2023. Development and validation of a genome-wide InDel marker set discriminating the alleles between the BB-genome Oryza species and rice (*O. sativa*). *Current Plant Biology*, *34*, p.100285.

Mo, Z., Lou, W., Zhang, Y., Hu, L., Zhai, M. and Xuan, J., 2024. Insertion/deletion variant characterization and marker development in pecan [*Carya illinoinensis* (Wangenh.) K. Koch]. *Scientia Horticulturae*, *325*, p.112660.

Rathi, S., Upadhyay, S., Singh, P.K., Kumar, R., Bisen, P., Loitongbam, B. and Snehi, S., 2021. Study of parental polymorphism and allelic variation for grain quality and yield traits in rice (*Oryza sativa* L.) using SSR markers. *International Journal of Plant & Soil Science*, *33*(24), pp.136-149.

Rathod, R., Suman, K., Madhubabu, P., Subbarao, L.V., Eswari, K.B., Chary, D.S. and Neeraja, C.N., 2021. Identification of genomic regions associated with grain zinc concentration in RILs derived from popular cultivar MTU1010 and BR2655 of rice. *Indian Journal of Genetics and Plant Breeding*, *81*(04), pp.538-547.

Shankar, A., Joseph, J., Francies, R.M. and Mathew, D., 2023. Evaluation for Parental Polymorphism and Identification of Microsatellites Linked to Drought Tolerance in Rice (*Oryza sativa* L.). *International Journal of Plant & Soil Science*, *35*(22), pp.275-284.

Shivani, D., Suman, K., Madhubabu, P., Rathod, R., Cheralu, C., Shankar, V.G. and Neeraja, C.N., 2020. Parental polymorphism in iron-and zinc-rich rice varieties (Swarna and type 3) Using ssr markers. *Applied Biological Research*, *22*(1), pp.69-75.

Singh, S.K., Majhi, P.K., Anandan, A., Korada, M., Habde, S.V., Khaire, A.R., Singh, D.K., Jayasudha, S., Singh, A. and Bhagvan, A.P., 2022. Microsatellites based parental polymorphism survey for moisture stress in Rice (*Oryza sativa* L.) between the parental genotypes HUR-1309 and CR Dhan 801. *Emergent Life Sciences Research*, *8*, pp.248-258.

Swamy, B.M., Kaladhar, K., Shobha Rani, N., Prasad, G.S.V., Viraktamath, B.C., Reddy, G.A. and Sarla, N., 2012. QTL analysis for grain quality traits in 2 BC_2F_2 populations derived

UNDER PEER REVIEW

from crosses between *Oryza sativa* cv Swarna and 2 accessions of *O. nivara*. *Journal of Heredity*, *103*(3), pp.442-452.

Vaughan, D.A. and Zhang, Q., 1997. Wild Rice: *Oryza rufipogon* and Its Importance in the Genus Oryza. In: Genetics and Genomics of Rice. Springer, pp.75-90.

Wang, R.R.C. and Li, Y., 2008. Genetic Diversity of Wild Rice *Oryza rufipogon*: A Study of Genetic Variation and Implications for Rice Improvement. Plant Breeding Reviews, 30, pp.1-24.

Welch, R.M. and Graham, R.D., 2004. Breeding for micronutrients in staple food crops from a human nutrition perspective. *Journal of experimental botany*, *55*(396), pp.353-364.

Yerva, S.R., Singh, S.K., Habde, D.S.S. and Behera, P.P., 2018. Parental polymorphic survey for high Zn and Fe content in grains of rice (*Oryza sativa*. L) using SSR markers. *Journal of Pharmacognosy and Phytochemistry*, *7*(4), pp.1362-1367.

Yu, J., Wang, J., Lin, W., Li, S., Li, H., Zhou, J., Ni, P., Dong, W., Hu, S., Zeng, C. and Zhang, J., 2005. The genomes of *Oryza sativa*: a history of duplications. *PLoS biology*, *3*(2), p.e38.

UNDER PEER REVIEW