

Genetic diversity of Indonesian rice varieties for salinity tolerance using RAPD markers

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Abstract. Nurhidayati T, Rachmandika FCA, Jundana MQ, Rahayu AE, Herwibawa B, Saputro TB. 2025. Genetic diversity of Indonesian rice varieties for salinity tolerance using RAPD markers. *Biodiversitas* 26: 731-738. Rice (*Oryza sativa*) is essential for food security in Indonesia, but agricultural land conversion and rising salinity threaten production. Therefore, it is crucial to study germplasm collections for salt tolerance and utilize molecular markers to select resistant lines. This study investigated the potential marker for salinity tolerance using local Indonesian rice. We investigated the genetic diversity of five germplasm collections (i.e., IR64, Jeliteng, Mantap, Pamerah, and Situ Bagendit) using a molecular marker technique with Random Amplified Polymorphic DNA (RAPD). The genomic DNA was extracted using CTAB 2%. This research employed ten RAPD primers (OPA-02, OPA-10, OPA-13, OPB-07, OPC-02, OPD-08, OPI-01, OPK-20, OPU-19, OPU-20). The analysis revealed a significant level of polymorphism, producing 184 bands, of which 168 were polymorphic, indicating 100% polymorphism; notably, the OPI-01 primer exhibited 94% polymorphism. The amplified bands ranged from 250 to 1920 base pairs, with the OPC-02 primer showing the most promising results, evidenced by a PIC value of 0.431. These findings suggest that the identified RAPD markers can effectively enhance the selection of salt-resistant rice varieties. This study highlights the potential of molecular markers, such as OPC-02, that could be valuable tools for selecting salt-tolerant rice varieties and contributing to food security amidst the challenges of agricultural land conversion and soil salinity.

Keywords: Genetic diversity, *Oryza sativa*, RAPD markers, salinity tolerance

INTRODUCTION

Rice (*Oryza sativa* L.) is a fundamental food source for millions of people globally, playing a crucial role in addressing hunger and food insecurity, particularly through initiatives aimed at developing climate-resilient varieties that support Sustainable Development Goal (SDG) number 2, targeting "zero hunger" (CGIAR 2017). In Indonesia, the fourth most populous country with 275.77 million people in 2022, food security remains a pressing issue (Rozaki 2021; BPS 2022). According to the report of Indonesian Food Security Index or in Indonesia well known as Indeks Ketahanan Pangan (IKP) in 2022, 74 districts and cities fell into the low IKP category, with significant agricultural land conversion exacerbating food insecurity (Tono 2022). From 2020 to 2022, the area of rice fields in Indonesia decreased by 204,602.96 hectares (BPS 2022), highlighting the need for intervention.

Rice production faces challenges from biotic and abiotic factors, notably salinity, which affects over 6% of the world's agricultural land (Ismail and Horie 2017; Yuvaraj et al. 2020). The study conducted by Singh et al. (2021) highlights the importance of understanding salinity tolerance mechanisms to support the development of salinity-tolerant varieties, which can enhance productivity in marginal lands. Salinity stress in rice can reduce growth and productivity through ionic stress, osmotic stress, and nutrient imbalance.

The absorption of Na⁺ and Cl⁻ ions can lower the osmotic potential between roots and soil solution, and reduce water infiltration availability, causing ion toxicity and decreasing nutrient uptake (Ullah et al. 2021). Salinity can also reduce endogenous phytohormone levels, hindering seedling formation, causing physiological disturbances due to reduced stomatal conductivity, and limiting the fixation and catalytic capacity of carbon enzymes (Ullah et al. 2019). Salinity is a critical issue for Indonesia, as the country is an agrarian nation vulnerable to the effects of soil salinity. Effective strategies to enhance rice production under salinity stress include rapid selection for superior varieties using molecular approaches that encompass the whole genome, allowing for the identification of genetic variants associated with salinity tolerance (Yuan et al. 2020).

Previous research by Mazumder et al. (2020) revealed significant polymorphism among local cultivars in Bangladesh. Random Amplified Polymorphic DNA (RAPD) primer has produced 255 polymorphic bands, showing 100% polymorphism. This study has demonstrated that RAPD primers effectively distinguish rice varieties based on DNA banding profiles. Another advantage of RAPD is its ability to reach most of the genome region and not require detailed information about the genome being studied (Babu et al. 2021). The selected local Indonesian rice varieties-Mantap, IR64, Jeliteng, Pamerah, and Situ Bagendit-are recognized for their yield potential and resilience to pests and diseases.

Molecular techniques can expedite the selection of salinity-tolerant rice varieties, with RAPD primers amplifying DNA fragments of various sizes, thus enhancing the detection of polymorphisms linked to salinity tolerance (Ghose et al. 2016).

A strategy to increase rice production under salinity stress conditions can be carried out with rapid selection to obtain superior varieties, namely through a molecular approach covering the whole genome or whole genome in rice plants. The choice of whole genome or whole genome is because it contains all the genetic information of an organism, including all genes, gene regulation, and non-coding elements that play a role in the genetic variation in a population. The whole genome allows the identification of various genetic variants, such as Insertion-Deletion Polymorphisms (InDels), which can play an adaptive role in the environment, including salinity tolerance (Yuan et al. 2020).

The local Indonesian rice varieties chosen for this study (Mantap, IR64, Jeliteng, Pamerah, and Situ Bagendit) were selected based on their designation as superior varieties by the Ministry of Agriculture of the Republic of Indonesia. It indicates that these local rice varieties have high yield potential, are resistance to pests and diseases, and have excellent taste. Moreover, using molecular approaches in rice research can accelerate the selection process of salinity-tolerant rice varieties. DNA analysis can be completed in days or weeks, compared to conventional methods that require months or years to grow, observe, and evaluate the agronomic traits of rice in the field. RAPD (Random Amplified Polymorphism DNA) primers were chosen because they have varying lengths and types of nitrogenous bases, amplifying DNA fragments of various sizes. It increases the likelihood of detecting DNA polymorphisms associated with salinity tolerance (Ghose et al. 2016). Therefore, similar research needs to be carried out on local rice in Indonesia, considering that Indonesia has a high biodiversity level. It shows that these local rice varieties have the potential for high yields, resistance to pests and diseases, and high flavor (Hairmansis et al. 2017; Paiman et al. 2020; Hidayanto et al. 2021). This research contributes to assessing the genetic variation of Indonesian local rice (*Oryza sativa*) using molecular markers related to salinity tolerance, determining the correlation between phenotypes and genetic diversity in salinity tolerance, and identifying potential RAPD molecular markers for salinity resistance in rice.

MATERIALS AND METHODS

Plant materials

The selected five local rice varieties were IR64, Jeliteng, Mantap, Pamerah, and Situ Bagendit. The Indonesian Rice Research Institute (IRRI), Subang, West Java, obtained local rice varieties. The growing medium used in this study was hydroponic. The hydroponic growing medium was prepared using water, nutrient A, and nutrient B in a ratio of 990 mL: 5 mL: 5 mL. This dosage is based on the usage instructions on the AB mix hydroponic Surabaya packaging, with nutrients A containing the macronutrients N, P, K, Ca, Mg, and S, and nutrient B containing the micronutrients Fe,

Mn, Zn, B, Cu, and Mo. Six plastic containers were used as hydroponic growing medium vessels. Each container was filled with a total solution volume of 10 L of 9.9 L of water and 50 mL each of nutrient A and B, then stirred until homogeneous. The hydroponic growing medium was periodically stirred to prevent nutrient sedimentation and to distribute nutrients within the medium (Nguyen et al. 2016).

Rice seedlings of uniform size were selected for planting. The selected rice seedlings were placed in sponges and positioned on an alvaboard that had been pre-drilled. The rice planting was carried out in six containers without salinity stress as an initial adaptation to the hydroponic growing medium. Salinity stress treatment was applied when the rice plants were 7 days after planting (DAP) in six containers containing 0 mM and 75 mM salinity concentrations.

DNA extraction

Equipment sterilization is carried out for each type of tool used in DNA isolation and extraction. These items are autoclaved at 121°C and 1.5 atm pressure for 60 minutes. Then, the equipment is oven-dried at 80°C for 2 hours. DNA extraction was performed using the modified CTAB 2% method of modification by Doyle and Doyle (Kouakou et al. 2022). The CTAB 2% composition was prepared by mixing 100 mL of 1 M Tris HCl pH 8, 280 mL of 1.4 M NaCl, and 40 mL of 0.5M EDTA in 1 L of distilled water. The mixture was stirred with a magnetic stirrer until homogeneous and stored at room temperature. The CIAA (chloroform and isoamyl alcohol) solution was prepared according to Busi et al. (2020) by mixing chloroform and isoamyl alcohol in a ratio of 24:1.

Rice seedlings at approximately 2-3 cm height were collected for DNA isolation. 10-20 mg of seedlings were cut and placed into 1.5 mL microtubes. 200 µL of 2% CTAB was added and ground with a pestle in liquid nitrogen until smooth. 300 µL of 2% CTAB was added and vortexed. The mixture was incubated in a Dry Block Heating Thermostat at 55°C for 30 minutes. 500 µL of chloroform isoamyl alcohol (CIAA) was added and mixed gently. The solution was centrifuged at 10,000 rpm for 10 minutes at 40°C. The supernatant was transferred to a new microtube. 200 µL of isopropanol was added to the new microtube, mixed gently, and incubated at 4°C for 2-3 hours. The mixture was centrifuged at 10,000 rpm for 5 minutes at 4°C. The supernatant was discarded, and 700 µL of cold 70% ethanol was added. The mixture was centrifuged at 10,000 rpm for 1 minute at 40°C. The supernatant was discarded. The pellet was dried. 50 µL of ddH₂O was added and stored at -200°C (Shu et al. 2018).

RAPD primer selection

Ten RAPD primers were selected for genetic variation analysis (Table 1). These primers, obtained from Integrated Primers at DNA Technologies, Inc. (Coralville, IA, U.S.A.), were selected based on their documented high degree of polymorphism as reported by previous studies (Kurup et al. 2009; Saputro et al. 2016). It is important to acknowledge that the polymorphism exhibited by RAPD primers can vary depending on the specific rice cultivars under investigation.

PCR amplification

The PCR amplification protocol employed RAPD primers and followed the general outline established by Ibitayo et al. (2017), with some adjustments made to the thermal cycling conditions. Each reaction was conducted in a 25 μ L volume containing 12.5 μ L of a pre-mixed Master Mix (Promega Go Taq® G2 Green Master Mix), 1.0 μ L of a RAPD primer, 2.0 μ L of the DNA template, and 7.5 μ L of nuclease-free water. A standard Bio-Rad Thermal Cycler was used for amplification. The thermal cycler was set to begin with a pre-denaturation step at 95°C for 3 minutes, followed by 35 cycles. Each amplification cycle incorporated a high-temperature denaturation step at 95°C for 20 seconds, followed by an annealing step lasting 1 minute at a temperature specific to the melting temperature (T_m) of the corresponding primer. It was followed by an extension phase at 72°C for 40 seconds. After completing 35 cycles, a final extension step was performed at 72°C for 10 minutes was performed, and the reaction mixture was held at 4°C to stabilize the amplified products. The RAPD-PCR amplified products were examined through electrophoresis on a 1% agarose gel, employing MiniPCR bio Gel Green™ Nucleic Acid Stain 10,000X in water and 1X TAE Buffer. The analysis used a Mini-Sub Cell GT Horizontal Electrophoresis System paired with a PowerPac Basic Power Supply #1640300 (voltage 100 V, for 50 minutes). The gel was subsequently illuminated under a UV light transilluminator.

Scoring of alleles

The stained gels were analyzed by evaluating the banding patterns in the photographs. The molecular weight of each amplicon, measured in base pairs, was estimated using ImageJ software. Gel electrophoresis was employed to separate the amplified DNA fragments (amplicons). A molecular weight marker, such as the Generuler™ 1kb DNA ladder, was used to estimate the size of each amplicon based on its migration pattern within the gel. The position of each band on the gel served as a unique identifier, with fragment sizes reported in base pairs (bp).

Data analysis

The values obtained from the matrix were utilized to compute the polymorphism information content (PIC) (Serrote et al. 2020). The calculation of polymorphism information content (PIC) value was performed using the following formula:

$$PIC = 1 - \sum P_{ij}^2$$

The symbol Σ signifies the total frequency of a specific primer across multiple alleles, where P_{ij} represents the frequency of the j th allele for primer i , with the summation extending over n alleles (Dwivedi et al. 2018). Cultivars were compared in pairs to generate a similarity matrix using the presence or absence of distinct and shared amplification results. The Relative mobility (RM), as defined by Semenov et al. (2023), was computed using this formula:

$$RM = \frac{\text{Migration distance of the band formed}}{\text{Migration ladder distance}}$$

Phylogenetic tree construction

The phylogenetic tree construction used R 4.2.2 (R Core Team 2022). Each DNA fragment amplified by a specific primer was considered a discrete genetic unit. The RAPD fragments were analyzed using a binary coding system, distinguishing their presence or absence and recording their presence as “1” or absence as “0” for each primer-cultivar combination. The phylogenetic tree construction used the Jaccard method (Zou et al. 2024).

RESULTS AND DISCUSSION

Effect of salt stress on local rice population

Figure 1 presents the results from the analysis of variance conducted on the rice population under salinity stress. Parameters including plant height (PH), total root length (RL), shoot fresh weight (SFW), root fresh weight (RFW), and shoot dry weight (RDW) demonstrated highly significant effects.

Table 1. The morphological assessment results of rice plants

Varieties	PH0 (cm)	PH75 (cm)	RL0 (cm)	RL75 (cm)	SFW0 (g)	SFW75 (g)	RFW0 (g)	RFW75 (g)	SDW0 (g)	SDW75 (g)
IR64	19.78	14.67	6.93	9.2	100.1	48.86	25.37	20.33	12.63	11.01
Mantap	18.29	14.82	6.9	7.86	111.16	70.86	23.86	19.63	14.41	11.1
Situ Bagendit	18.08	16.38	6.86	9.36	97.2	47.5	20.46	18.13	15.04	13.27
Pamerah	20.49	10.78	6.53	5.38	103.13	46.63	26.13	19.63	19.09	8.56
Jeliteng	18.78	12.67	6	6.54	101.93	37.16	20.73	15	17.61	9.67

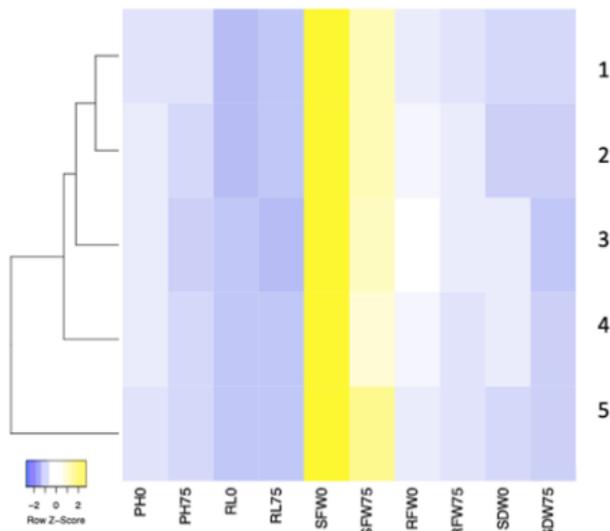


Figure 1. Heatmap related to 5 local and improved cultivars of Indonesian rice phenotypes. 1: Situ Bagendit, 2: IR64, 3: Pamerah, 4: Jeliteng, 5: Mantap. PH: Plant height (cm), RL: Root length (cm), SFW: Shoot fresh weight (g), RFW: Root fresh weight, SDW: Shoot dry weight (g). Measurements at 0 mM (control) are labeled as PH0, RL0, SFW0, RFW0, and SDW0, while those at 75 mM salt concentration are labeled as PH75, RL75, SFW75, RFW75, and SDW75.

To address the salinity tolerance levels of the rice varieties used in this study, Figure 1 illustrates the distinct responses of five varieties (Situ Bagendit, IR64, Pamerah, Jeliteng, and Mantap) under control (0 mM) and salinity stress (75 mM). The heatmap shows Z-scores representing how each phenotype deviates from the mean, where yellow indicates a positive Z-score (above the mean) and blue indicates a negative Z-score (below the mean). Mantap consistently demonstrates superior performance under salinity stress, with positive Z-scores for plant height (PH), root length (RL), shoot fresh weight (SFW), root fresh weight (RFW), and shoot dry weight (SDW). It indicates that Mantap is highly tolerant to salinity, supported by its strong phenotypic traits under stress. Conversely, IR64 shows predominantly negative Z-scores, highlighting its sensitivity to salinity stress. The results from Situ Bagendit, Jeliteng,

and Pamerah exhibit intermediate tolerance, as their Z-scores for various parameters range closer to the mean.

To further clarify, the Mantap variety is a recently released cultivar with documented resilience to environmental stresses. It is characterized by a high potential yield (up to 9.1 tonnes/ha) and moderate disease resistance, as supported by the Minister of Agriculture Decree 81/HK.540/C/02/2019. While this study lacks published references specifically classifying the salinity tolerance of all varieties, the observed phenotypic responses under controlled conditions provide reliable indicators of their tolerance levels. Therefore, this study uses Mantap as a highly tolerant reference variety and IR64 as a sensitive baseline for identifying RAPD markers associated with salinity tolerance.

Molecular marker related to salinity stress

The present study uncovers a remarkable degree of genetic heterogeneity within the selected varieties and local landraces, underlining the importance of preserving this genetic diversity. The data presented in Table 2 demonstrates that all primers produced polymorphic locus (100% polymorphic locus percentage for all primers). Additionally, OPU-19 yielded the highest number of polymorphic loci (54), whereas OPK-20 generated the fewest polymorphic loci (2). Based on these data, the PIC values ranged between 0.297 (OPI-01) and 0.431 (OPC-02). Primer OPC-02 exhibited the highest PIC value, indicating high genetic diversity at the loci amplified by this primer. Conversely, primer OPI-01 possessed the lowest PIC value, suggesting low genetic diversity at the loci amplified by this primer.

The RAPD-PCR was used to develop molecular markers for salinity tolerance with 10 primers. Nine of these primers successfully amplified DNA fragments across all genotypes, generating between 2 and 34 bands. Primer OPC-02 revealed a potential molecular marker for salinity tolerance by showing a band of 1069 bp in the salt-tolerant Mantap population, suggesting it a positive marker for salinity tolerance in rice. Conversely, bands of 554 bp and 727 bp were absent in the salt-tolerant Mantap population, indicating these as potential negative markers for salinity tolerance in rice plants.

Table 2. Details of RAPD primers, including the number of associated bands, size ranges, polymorphic bands, and PIC values

Name of primers	Range of band sizes (base pairs)	Sequence	Tm (°C)	Total locus	Total locus polymorphic	Percentage locus polymorphic (%)	PIC value
OPA-O2	950-1900	5'-TGCCGAGCTG-3'	40.7	4	4	100	0.320
OPA-10	530-1920	5'-GTGATCGCAG-3'	33.1	26	26	100	0.395
OPA-13	890-1630	5'-CAGCACCCAC-3'	37.7	10	10	100	0.338
OPB-07	860-1700	5'-GGTGACGCAG-3'	38.1	22	22	100	0.393
OPC-02	330-1300	5'-GTGAGGCGTC-3'	37.6	25	25	100	0.431
OPD-08	330-1150	5'-GTGTGCCCCA-3'	40.1	11	11	100	0.380
OPI-01	900-1930	5'-ACCTGGACAC-3'	33.4	16	15	94	0.297
OPK-20	1300-1400	5'-GTGTCGCGAG-3'	38.5	2	2	100	0.320
OPU-19	250-1620	5'-GTCAGTGCGG-3'	38.1	54	54	100	0.391
OPU-20	360-1580	5'-ACAGCCCCCA-3'	41.5	14	14	100	0.391

Table 3. The pairwise inter-variety similarity index (S_{ij}) among 5 local and improved varieties of Indonesian rice genotypes

Genotype	A	B	C	D	E
A	1.00000000				
B	0.50594694	1.00000000			
C	0.02378774	0.00000000	1.00000000		
D	0.26052150	0.2367338	0.008462946	1.00000000	
E	0.26738335	0.2435956	0.015324794	0.407136322	1.00000000

Notes: A: IR64, B: Jeliteng, C: Mantap, D: Pamerah, E: Situ Bagendit

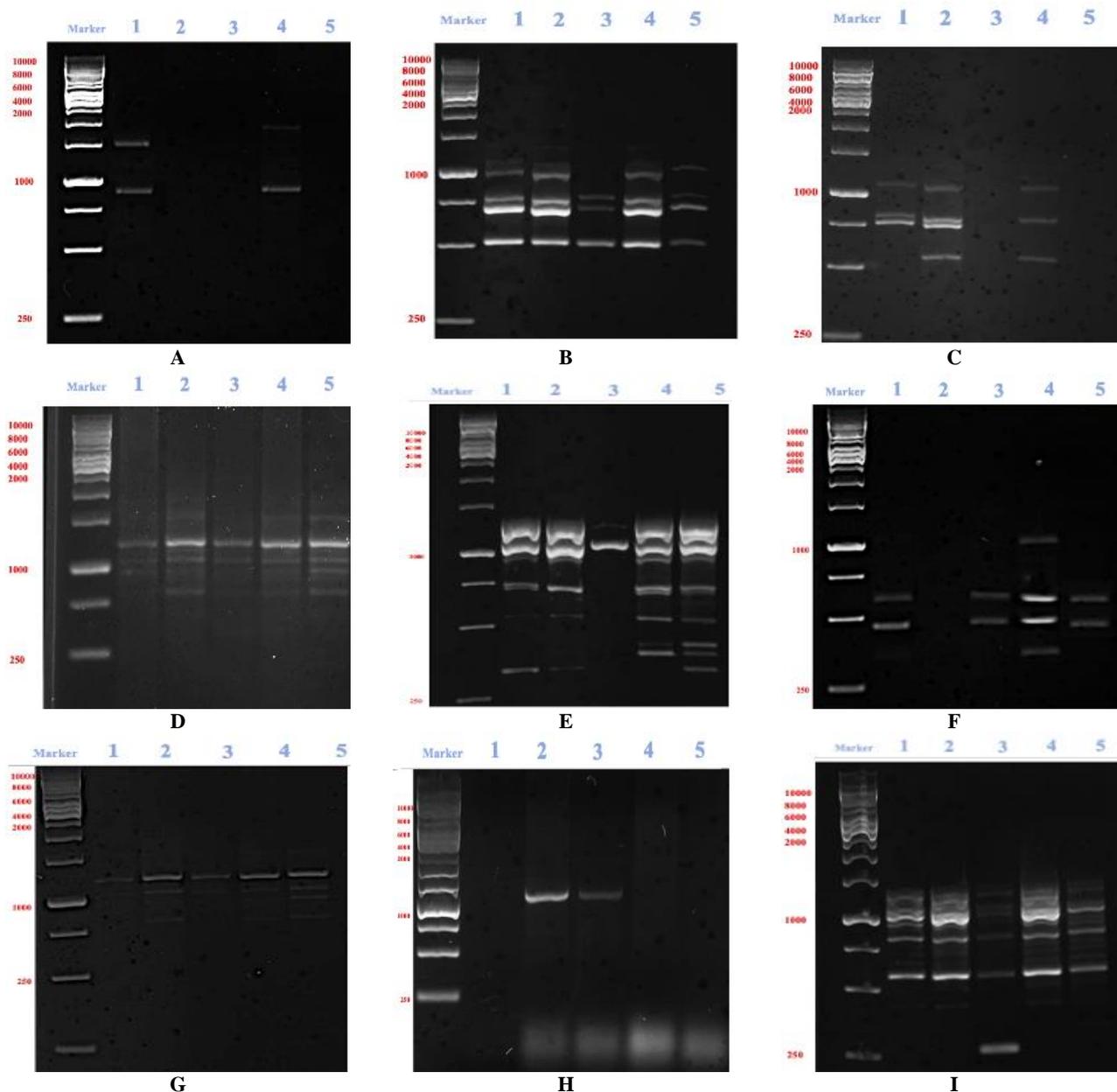


Figure 2. RAPD banding pattern of 5 local and improved varieties of Indonesian rice generated by 10 random amplified primers. A. OPA-02; B. OPA-10; C. OPA-13; D. OPB-07; E. OPC-02; F. OPD-08; G. OPI-01; H. OPU-19; I. OPK-2

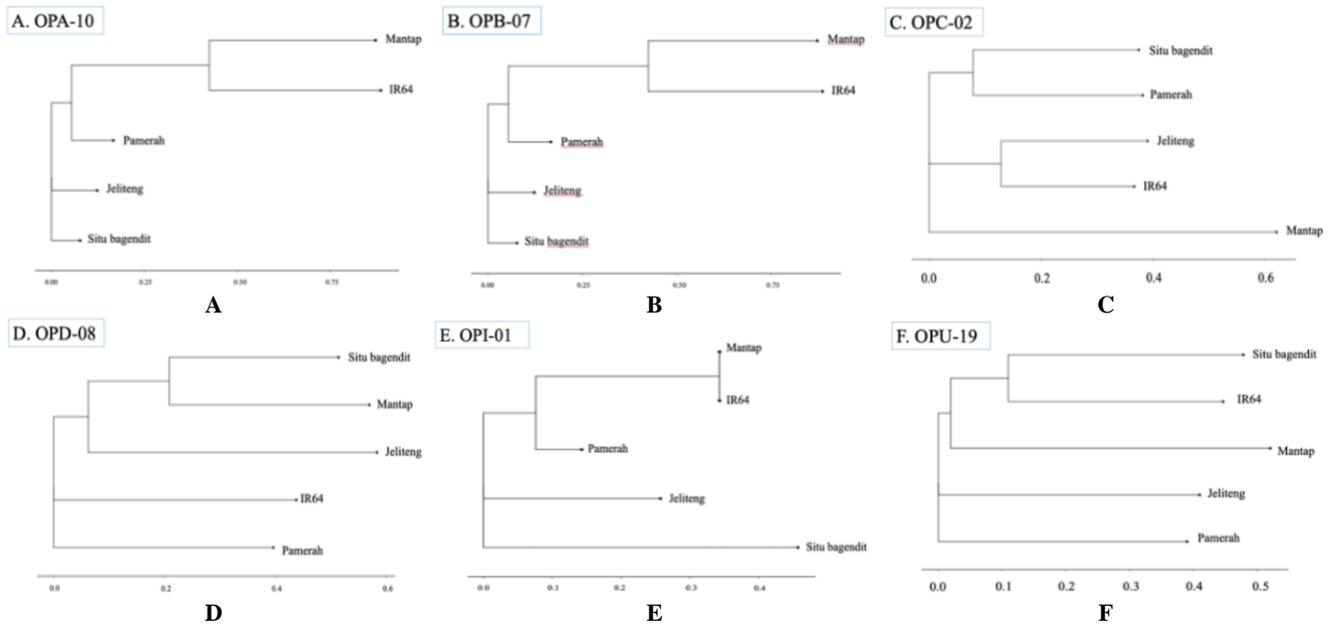


Figure 3. Phylogenetic tree of local Indonesian rice varieties (Varieties IR64, Jeliteng, Mantap, Pamerah, Situ Bagendit). A. RAPD Primer OPA-10; B. RAPD Primer OPB-07; C. RAPD Primer OPC-02; D. RAPD Primer OPD-08; E. RAPD Primer OPI-01; F. RAPD Primer OPU-19

Polymorphism analysis of RAPD markers

This experiment aims to understand the genetic variation of local Indonesian rice plants (*O. sativa*) based on molecular markers associated with their salt tolerance levels. These PIC values obtained in this investigation reveal that all these primers employed were highly informative and efficient in distinguishing genotypes. Molecular markers can serve as highly effective indicators in molecular phylogeny studies, genetic diversity analyses, and genetic identification, including selecting plant materials and varieties (Nath et al. 2018).

This study selected Random Amplified Polymorphic DNA (RAPD) primers due to their advantages as molecular markers for salinity tolerance in rice, particularly in the initial research stages. RAPD exhibits high polymorphism, producing numerous unique DNA bands that can be easily detected, allowing for the identification of genetic variations associated with salinity tolerance with high sensitivity (Bousba et al. 2020). RAPD is relatively simple and inexpensive compared to other molecular marker techniques. The RAPD procedure does not require complex laboratory equipment, making it easy for researchers to use at various levels. RAPD can directly detect DNA polymorphisms without requiring nucleotide sequence information (Kumari and Takur 2014). RAPD enables the analysis of molecular markers in rice varieties without annotated genomes, a typical case for many local rice varieties. Many local rice varieties have high genetic variation, which can make genome annotation more difficult. Additionally, research on local rice often focuses on developing new varieties with desired traits, such as resistance to pests or diseases (Probojati et al. 2019). RAPD allows simultaneous multi-locus analysis to detect polymorphisms at several loci in a single reaction. It increases the efficiency of the analysis

and enables the more comprehensive identification of molecular markers associated with salinity tolerance (Younis et al. 2020).

Based on Figure 2, the gel electrophoresis results show several bands not formed by the rice varieties and RAPD primers. The lack of amplification observed with specific RAPD primers can be attributed to two primary factors: firstly, the absence of complementary sequences within the genomic DNA, and secondly, the presence of only one DNA strand containing complementary sequences that align with the primer (Amiteye 2021). Additionally, factors influencing the PCR amplification process include annealing temperature. Screening or optimizing PCR is crucial for generating informative band patterns related to DNA polymorphism. Optimization focuses on adjusting DNA denaturation and annealing temperatures in the PCR machine. Low denaturation temperatures can hinder the separation of DNA double strands, thereby impeding the process of DNA polymerization (Kadri 2019).

To achieve DNA amplification with a single random primer, the primer must have a base sequence complementary to both strands of the genomic DNA at opposite positions (Wang et al. 2022). A considerable distance between complementary pairs of the DNA template will prevent amplification. To achieve optimal PCR-RAPD results, selecting appropriate DNA binding sites and ensuring high-quality DNA templates is necessary. The purity and integrity of the DNA template can be assessed based on the purity and size of the DNA genome. DNA contamination, such as by secondary metabolites (phenols), can interfere with DNA amplification (Putra et al. 2020).

Based on Figure 3.C, it can be observed that the RAPD primer capable of distinguishing the Mantap variety from the other four varieties is the OPC-02 primer. No other

primer demonstrates a similar ability to the OPC-02 primer. OPC-02 primer has demonstrated utility as a molecular marker associated with salinity stress in rice. It aligns with the findings of Saputro et al. (2016), who suggested that RAPD primer OPC-02 could serve as a molecular marker for salinity stress in maize callus growth. Additionally, research conducted by Kurup et al. (2009) also indicated primer OPC-02 could be a molecular marker for salinity tolerance in date palm plants.

Correlation between phenotype and genetic diversity of salinity tolerance in rice

Investigating the correlation between phenotype and genetic diversity of salt tolerance in rice (*O. sativa*). This level of similarity is typically measured using sequence identity percentage or genetic distance. The higher the value, the greater the similarity between the two varieties (Bajusz et al. 2021). Table 3 represents the similarity index of local Indonesian rice varieties, illustrating the level of similarity between two or more compared varieties. This similarity level is typically measured using the percentage of sequence identity or genetic distance. The higher the value, the greater the similarity between the two varieties (Ilham et al. 2022). The highest relatedness is between the varieties Jeliteng and IR64, with a similarity index of 0.5059469. The varieties Mantap and Pamerah exhibit a similarity index of 0.008462946.

In conclusion, the extensive genetic diversity observed among the selected genotypes underscores the utility and efficacy of RAPD markers in facilitating comprehensive diversity analysis studies. This study found that the OPC-02 primer was the most effective and informative primer to be used as a candidate molecular marker. Genetic variation can facilitate maintaining developmental stability and maximizing biological potential in organisms. This experiment unveiled notable variability among five local Indonesian rice varieties, highlighting the rich genetic diversity of rice in Indonesia. The data presented in this study offer valuable resources for researchers developing rice breeding programs. This information can inform decisions regarding selective breeding, crossbreeding, and mutation breeding strategies, ultimately contributing to enhanced rice cultivation practices. It can yield new rice varieties characterized by enhanced salinity tolerance and increased yield potential. Additionally, previous research found that the Mantap variety has high salinity tolerance and adaptability to new environments. Therefore, a linear correlation between phenotypes and genetic diversity in salinity tolerance in rice (*O. sativa*). Additional research is necessary to map and identify the levels of salinity tolerance in the rice genotypes employed in this study.

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