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Morpho-Molecular Diversity Analysis of Local Rice (*Oryza sativa* L.) Genotypes Using Microsatellite Markers

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Authors' contributions

This work was carried out in collaboration among all the authors. Authors SKS, CS and MK designed the study and wrote the protocol. Author CS performed statistical analysis and wrote the first draft of manuscript. Authors MK, SH and DKS managed the further analyses of the study and improved the manuscript. Authors AK and PKM managed the literature searches. All authors read and approved the final manuscript.

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ABSTRACT

Aim: The knowledge of genetic diversity and relationship among the genotypes play a significant role for genetic enhancement in breeding programmes to increase production, improve quality, biotic and abiotic stresses, and also for the selection of superior parental lines in rice. The present field experiment was conducted to study the diversity present in 29 local genotypes of rice using both morphological and molecular ways.

Methodology: The experiment was conducted at Agricultural Research Farm, Banaras Hindu University, during *Kharif*-2017 in an augmented block design with 29 rice genotypes including 3 checks. Mahalanobis' D^2 analysis was carried out to assess the morphological diversity present among the genotypes and molecular analysis was done with 21 polymorphic SSR markers using the NTSYSpc software.

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Results: Mahalanobis' D^2 grouped the 29 genotypes into 6 clusters based on the *inter-se* genetic distance. The highest intra-cluster distance was recorded in the Cluster I (32.73), which comprised of 7 genotypes. The highest inter-cluster distance (65.86) was observed between Clusters IV and V. Molecular diversity analysis grouped the 29 rice genotypes into 2 main clusters *i.e*. cluster I and cluster II with dissimilarity coefficient of 0.34, which were further divided into sub-clusters. Polymorphic Information Content (PIC) value is an evidence of diversity and frequency among the varieties. The level of polymorphism varied from 0.164 to 0.694, with an average 0.521. The highest PIC value was observed for locus RM 5 (0.694) followed by RM 510 (0.692). All the 21 primers showed polymorphism and the number of alleles ranged from 2 to 4 with an average of 3.04.

Conclusion: This study established the presence of considerable amount of genetic diversity among the genotypes studied, the most diverse genotypes being Anupam gold and HUR-1309 followed by Kalanamak-2 and HUR-1304. Breeders may attempt hybridization among the above genotypes which showed maximum diversity, for creating more variability in rice and can be used for planning further breeding programmes.

Keywords: D^2 analysis; dendrogram; molecular diversity; Oryza sativa; Polymorphic Information Content (PIC); microsatellite marker.

1. INTRODUCTION

Rice (*Oryza sativa* L.) belongs to the genus *Oryza* of family Poaceae. The genus *Oryza* is known to consist of 2 cultivated species *i.e.,* Asian rice (*Oryza sativa* L. 2n=24) and African rice (*Oryza glaberrima* S. 2n=24) and 22 wild species (2n=24, 48) (Jena KK, 2010) [1]. Asian rice has three distinct eco-geographical subspecies *viz.,* indica, japonica and javanica [2]. It is a diverse crop due to its adaptation to different geographical, ecological and climatic regions. It is a very important food crop of Asia and more than half of the world's population depends on rice for their dietary intake (FAO 2004) [3]. It is an essential food crop of the world both in terms of area and production. With an estimated increases in world population, the global agriculture production should meet the food demands of the increasing population. The Food and Agricultural organization of the United Nations (FAO) forecasts that the global food production will need to increase by 70% if the population reaches 9.1bn by 2050 [4]. Rice being a major crop in the world, plays an important role and increase in the production of rice depends on the development of improved varieties with better productivity and adaptability, which largely depends on the existence of sufficient genetic diversity in rice germplasm [5].

Assessing the genetic diversity is the prerequisite for any crop improvement programme as it helps in the development of superior recombinants. Genetic divergence among the genotypes plays an important role in finding of parents with huge variation for different characters [6]. Genetic diversity can be evaluated with morphological traits, seed proteins, iso-enzymes and DNA markers. Morphological characteristics might themselves be not enough to distinguish between pairs of closely related species, geographical races, or ecotypes, because not all genetic differentiation results in morphological differences. Thus a genetic characterization of natural resources is a critical step for a better understanding of genetic resources for the implementation of *in situ* and *ex situ* conservation activities and their utilization in future breeding programmes. For the evaluation of genetic diversity, molecular markers have been generally superior to morphological, pedigree, heterosis and biochemical data [7]. And these molecular markers also provide a reliable estimate of genetic diversity, may help in improving the showing efficiency for many traits through their linkage having alleles with small and large effects for quantitative and qualitative traits respectively [8]. Genetic diversity is usually measured by genetic distance or genetic similarity, both of which involve that there are either differences or similarities at the genetic level.

D^2-statistics of Mahalanobis (1928) is one of the most effective tools to measure the genetic distance between genotypes as measured by allelic frequencies at a sample of loci. This technique was found to be successful tool in quantifying the degree of divergence in the germplasm. In the same way, molecular marker technology provides an important tool in the assessment of genetic relationships within and among species, in which differences among accessions can be discovered at the DNA level [9,10]. Out of the several PCR-based markers use in genetic mapping studies, microsatellite markers, also known as simple sequence

repeats (SSR) markers are better because they are highly informative, co-dominant in nature, highly reproducible, abundant, easily analysed and cost effective [11]. These SSR markers are particularly used in evaluation of genetic similarity and dissimilarity among closely related individuals, plant species or populations [12]. Other applications of SSR markers in genomic analysis include, characterizations of genetic resources to support gene bank organizations [13], to estimate the genetic variability in genotypes used in breeding programmes etc. With this available background information, the present study was conducted with 29 local genotypes of rice.

2. MATERIALS AND METHODS

2.1 Plant Material and Design

The plant material for the present research work includes 29 local rice genotypes (including 3 checks). Detailed list of genotypes used in the current study is presented in Table 1. The evaluation took place at the Agricultural Research Farm, Banaras Hindu University, Varanasi, Uttar Pradesh, India in an augmented design during *Kharif*-2017. The 29 genotypes were raised on nursery bed and appropriate agronomic practices were done in the nursery. 21 days old seedling were transplanted in the main field at the standard spacing of 20 x 15 cm in 4 m rows in 3 rows each in augmented block design with 3 blocks having repeated checks. All the agronomic practices were carried out in the main field to grow a healthy crop.

2.2 Morphological Observations

Observations on yield and yield attributing traits were recorded on 5 randomly selected plants of each genotype in respective block. 16 quantitative characters viz., Days to first flowering, Days to 50% flowering, Plant height (cm), Panicle length (cm), Number of tillers/plant, fertile spikelets, sterile spikelets, Kernel length (mm), Kernel breadth (mm), Kernel L/B ratio, 1000 grain weight (g), Total grains/Panicle. Plot yield (kg), Grain yield (kg/ha), Biomass (kg/ha) and Harvest index (%) were recorded during different growth stages of the crop. Days to first flowering, Days to 50% flowering and Plot yield were recorded on plot basis whereas data for all other traits was recorded on five randomly selected plants. Mean of these five plants data was used for further analysis.

2.3 Molecular Diversity Analysis

A total of 21 SSR markers were selected on the basis of polymorphism shown by markers in screening (work not published). List of SSRs used in this study along with their details were presented in Table 2.

2.4 DNA Isolation to Scoring of DNA Bands

Young leaves were collected from 12-14 days old rice seedlings and the DNA was extracted following the CTAB extraction method according to Doyle and Doyle, 1987 [14] with few modifications. The DNA quality estimation was done using Biophotometer plus. The ratio (OD260/OD280 ratio) thus obtained was used to estimate the nucleic acid purity in the different DNA samples. Polymerase chain reaction (PCR) was performed to selectively amplify *in vitro* a specific selected segment of the total genomic DNA to a billion fold [15]. All the amplifications were performed in the Eppendorf Thermo-cycler (USA). All 21 primers were used to amplify the 29 genotypes and the amplified DNA products generated through SSR primers were resolved through electrophoresis in 2.5 per cent agarose gel prepared in TAE buffer [242 g Tris-base, 57.1 3ml glacial acetic acid and 100 ml 0.5 M EDTA (pH 8.0) dissolved in distilled water and final volume made to 1000 ml]. For electrophoresis, 15µl of the PCR product was mixed with 2µl of 6X loading dye (bromophenol blue) and loaded in the slot of agarose gel. In order to determine the molecular size of amplified products, the gels were loaded with 1µg of a 50 bp DNA marker (Fermentas, USA). Gel electrophoresis was carried out at a constant voltage of 65 V for about 3.5 hours and the gels were visualized under a UV light source in a gel documentation system (Gel DocTM XR+, BIO-RAD, USA). Finally, the images of amplification products obtained from all the 21 primers were scored for polymorphism for evaluation of diversity among the genotypes.

2.5 Statistical Analysis

2.5.1 Field screening: Analysis of Variance (ANOVA)

Quantitative trait mean values computed based on data on five randomly tagged plants in each genotype and check entry were used for statistical analysis. The trait means of each of the genotype was estimated [16] and this was used for all the statistical analysis.

2.5.2 Mahalanobis' D^2-statisticsand grouping of genotypes by tocher's method

In this investigation genetic divergence was estimated based on Mahalanobis' generalized distance as described by [17]. Original variable means were transformed to un-correlated variables by the pivotal condensation method of inversion matrix. The D^2-values between the genotypes were obtained as the sum of squares of differences of the values of the corresponding transformed variables. After arranging the D^2-values of all combinations of one genotype with the others in ascending order of magnitudes, the genotypes were grouped into a number of clusters by Tocher's method described by [17].

2.5.3 Molecular diversity

Molecular Marker based Genetic Diversity Analysis (MMGDA) also potential for assessing changes in genetic diversity over time and space [18]. Band position in comparative SSR profile for each genotype and primer combination was scored from the respective gel images. SSR profile from only genotype×primer combination , which gave constant amplification for all the genotype and without any blank lane per unclear bands, was included in this study. The amplified fragments were scored as '1' for the presence and '0' for the absence of a band generating the 0 and 1 matrix. These binary data matrix was then utilized to generate genetic similarity data among the 29 rice genotypes.

The effects of different scales of measurement for different quantitative traits were minimized by standardizing the data for each trait separately prior to cluster analysis. Standardization was done by dividing the deviation of mean for a line from the mean for 29 genotype with the standard deviation for the given trait; the STAND module of NTSYS [19] software was used to furnish the same.

2.5.4 Genetic dissimilarity and cluster analysis based on UPGMA

The binary data matrix generated by polymorphic SSR markers were subjected to further analysis using NTSYS-pc version 2.11W [19]. Jackard's dissimilarity coefficient was calculated using the SIMQUAL programme. The dissimilarity matrix was used as an input for analysis of clusters. UPGMA-based clustering was done using SAHN module of NTSYSpc for dendrogram construction. In Unweighted pair-group average (UPGMA) clusters are joined based on the average distance between all members in the two groups.

The Polymorphic Information Content (PIC) for SSR markers was calculated as per the formula,

$$PIC=1-(P_i2)$$

Where, '*i*' is the total number of alleles detected for each SSR marker, 'P_i' is the frequency of the i^{th} plus allele in the set of 29 genotypes studied. PIC value is used to estimate the discriminatory power of the SSR marker.

3. RESULTS AND DISCUSSION

The morphological and molecular data on preferences of the genotypes were analyzed to find out the variation among genotypes for different characters. The experimental results obtained from the present study conducted during *Kharif* 2017 for 29 local rice genotypes are presented in the following sub-heads.

Table 1. List of genotypes used in the present study (MTU-7029, HUR 105 and Rajendra Kasturi are the check genotypes)

1.HUR-1304	11.BHULC-9	21.HUR105
2. Kudrat -3	12.Golden Hira	22.Rajendra Kasturi
3.BHULC-7	13.BHULC-56	23.Golden basmati-2
4.Badshah Bhog	14.Garima	24.Sugandh- 100
5.HUR- 1309	15.GR- 32	25.HUR- 1308
6.Kalanamak -2	16.Kalanamak(original)	26.Pusa Basmati 1121
7.Anupam Gold	17.Kalanamak – 3	27.Vishnu Bhog white
8.BHULC-8	18.Golden G. R. 32 black	28.Jallahari
9.HUBR 2-1	19. MTU7029	29. Kudrat -1
10.Kalanamak(farmer)	20.NDR 8002	

Table 2. Details of the microsatellite markers used in present study along with No. of alleles and PIC value obtained after analysis

S. no.	Microsatellite locus	Forward/ Reverse	Sequence	T_m	No. of alleles	PIC value*
1.	RM 514	Forward	AGATTCATCTCCCATTCCCC CACGAGCATATTACTAGTGG	52	4	0.629
		Reverse		50		
2.	RM 5	Forward	TGCAACTTCTAGCTGCTCGA	52	4	0.694
		Reverse	GCATCCGATCTTGATGGG	51		
3.	RM 433	Forward	TGCGCTGAACTAAACACAGC	52	3	0.536
		Reverse	AGACAAACCTGGCCATTCAC	52		
4.	RM 277	Forward	CAAATCCCGACTGCTGTCC	51	4	0.667
		Reverse	TGGGAAGAGGAGAGCACTACAGC	52		
5.	RM 171	Forward	AACGCGAGGACACGTACTTAC	54	3	0.580
		Reverse	ACGAGATACGTACGCCTTTG	52		
6.	RM 284	Forward	ATCTCTGATACTCCATCCATCC	53	3	0.558
		Reverse	CCTGTACGTTGATCCGAAGC	54		
7.	RM 334	Forward	GTTCAGTGTTCAGTGCCACC	54	3	0.558
		Reverse	GACTTTGATCTTTGGTGGACG	52		
8.	RM 44	Forward	ACGGCAATCCGAACAACC	53	2	0.370
		Reverse	TCGGGAAACCTACCCTACC	54		
9.	RM 536	Forward	TCTCTCCTCTTGTTTGGCTC	52	3	0.440
		Reverse	ACACACCAACACGACCACAC	54		
10.	RM 552	Forward	CGCAGTTGTGGATTTCAGTG	52	4	0.673
		Reverse	TGCTCAACGTTTGACTGTCC	52		
11.	RM 431	Forward	TCCTGCGAACTGAAGAGTTG	55	3	0.591
		Reverse	AGAGCAAAACCCTGGTTCAC	55		
12.	RM 489	Forward	ACTTGAGACGATCGGACACC	54	2	0.365
		Reverse	TCACCCATGGATGTTGTCAG	52		
13.	RM 259	Forward	TGGAGTTTGAGAGGAGGG CTTGTTGCATGGTGCCATGT	50	4	0.660
		Reverse		52		
14.	RM 510	Forward	AACCGGATTAGTTTCTCGCC TGAGGACGACGAGCAGATTC	52	4	0.692
		Reverse		54		

S. no.	Microsatellite locus	Forward/ Reverse	Sequence	T_m	No. of alleles	PIC value*
15.	RM 215	Forward	CAAAATGGAGCAGCAAGAGC TGAGCACCTCCTTCTCTGTAG	52	3	0.583
		Reverse		54		
16.	RM 257	Forward	CCGTGCAACTTAAATCCAAACAGG	55	3	0.586
		Reverse	GGAATCCTATATGAGCCAGTGATGG	55		
17.	RM 536	Forward	TACCAGGATCATGTTTCTCTCC	52	3	0.164
		Reverse	ACTGTGAGATTGACTGACAGTGG	54		
18.	RM11943	Forward	CTTGTTCGAGGACGAAGATAGGG	55	2	0.335
		Reverse	CCAGTTTACCAGGGTCGAAACC	55		
19.	RM 11	Forward	TCTCCTCTTCCCCCGATC ATAGCGGGCGAGGCTTAG	53	3	0.573
		Reverse		53		
20.	RM 60	Forward	AGTCCCATGTTCCACTTCCG	54	2	0.361
		Reverse	ATGGCTACTGCCTGTACTAC	52		
21.	RM 474	Forward	AAGATGTACGGGTGGCATTC TATGAGCTGGTGAGCAATGC	52	2	0.346
		Reverse		52		

**PIC value: Polymorphic information content obtained after analysis*

3.1 Analysis of Variance (ANOVA)

The analysis of 16 traits was carried out to partition the total variation into genotypic variation and variation due to other sources. Analysis of variance was based on the mean values of 16 quantitative traits in 29 rice genotypes.

3.2 Genetic Diversity by Mahalanobis' D^2 Statistic

D^2 statistic has grouped the 29 genotypes including 3 checks into 6 different clusters based on the *inter se* genetic distances indicating presence of considerable diversity in the set of material under study. The composition of different clusters obtained from the D^2 analysis has been presented in the Table 3. The six clusters, I,II,III,IV,V and VI contained 7, 2, 3, 7, 5 and 5 genotypes respectively. The maximum number of genotypes was recorded in cluster-I and IV *i.e* 7. The clustering pattern reflected by the presence of considerable extent of genetic diversity in the material under study. A study by [20] using Mahalnobis' D^2, grouped 29 rice genotypes into 6 clusters. In the present grouping by Tocher method, maximum intra cluster distance was recorded in cluster-I (32.73) and minimum was recorded in cluster-V (13.97).

The intra and inter-cluster distances *i.e.* D^2-values have been presented in the Table 4 and the diagrammatic representation of clusters with intra and inter cluster D^2 values has been presented in Fig 1. It is said that genotypes grouped in one cluster are less divergent than those which are placed in different clusters. The highest inter-cluster distance was observed between the clusters IV and V (65.862) followed by clusters IV and VI (51.501) followed by clusters I and IV (47.79) indicating that the hybridization between these diverse genotypes would yield desirable segregants with the accumulation of favorable genes in the segregating generations. Similar results were also reported by [21,22,23]

3.3 Molecular Diversity Using SSR Primers

In the present investigation, study of diversity among 29 local rice genotypes was performed using 21 SSR markers. All the 21 SSRs showed polymorphism and scoring of alleles was done accordingly and this data is used for further analysis.

Table 3. Grouping of twenty-nine genotypes of rice into six clusters by Tocher Method (MTU-7029, HUR 105 and Rajendra Kasturi are the check genotypes)

S. no.	Cluster name	Number of genotypes	Name of genotypes
1.	Cluster I	7	Kudrat-1, HUR-1304, Kudrat-3, BHULC-7, BHULC-8, HUBR 2-1, HUR-1308
2.	Cluster II	2	Kalanamak-2, MTU7029
3.	Cluster III	3	HUR-1309, Golden Hira, NDR 8002
4.	Cluster IV	7	BHULC-56, Garima, GR-32, Kalanamak (original), Golden G.R.32 black, Golden basmati-2, Pusa basmati 1121
5.	Cluster V	5	Vishnu Bhog white, Badshah Bhog, Anupam Gold, Kalanamak, Kalanamak-3
6.	Cluster VI	5	Jallahari, BHULC-9, Sugandh-100, HUR105, Rajendra kasturi

Table 4. Average intra and inter Cluster distance D2 values among six clusters of 29 genotypes of rice by Tocher Method

	I cluster	II cluster	III cluster	IV cluster	V cluster	VI cluster
I Cluster	32.731	33.767	34.16	47.79	37.906	38.861
II Cluster		19.49	24.531	36.626	41.1	33.992
III Cluster			18.55	35.554	37.231	32.875
IV Cluster				14.861	65.862	51.501
V Cluster					13.968	31.567
VI Cluster						19.597

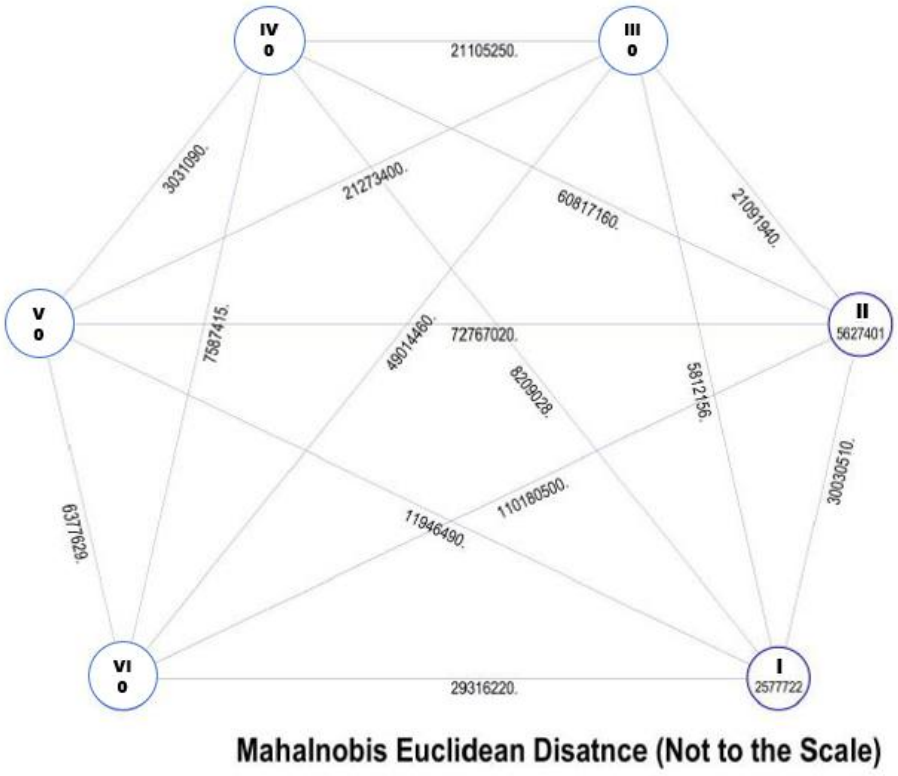

Fig. 1. Relation Disposition of cluster showing average genetic distance (D) between and within

3.3.1 Scoring of SSR band and PIC value

The polymorphic information content (PIC) was employed for each locus to evaluate the information of each marker and its discriminatory ability. PIC value of each marker can also be evaluated on the basis of all SSR loci and alleles [24]. In the current study the level of polymorphism was high and varied from 0.164 to 0.694, with an average 0.521. The highest PIC value was observed for locus RM 5 (0.694) followed by RM 510 (0.692) and RM 552 (0.673) and lowest by RM 536 (0.164). All the 21 primers showed polymorphism and the number of alleles ranged from 2 to 4 with an average of 3.04 (Table 2). In another study conducted by [25], they recorded PIC value ranging from 0.215 to 0.791 with an average of 0.25. Similar results were also obtained by [26] and [27]. Since PIC values higher than 0.50 indicate high polymorphism (Ahn *et al.,* 2018), this confirms that SSR markers used in the study were highly informative. The PIC values which were observed in the study were found comparable to previous estimates of microsatellite marker analysis in rice [28]. The given PIC values were found higher representing that genetic diversity in the present compilation was also higher [28]. A total of 64 microsatellites alleles were amplified from 21 markers and this demonstrates considerable variability among the genotypes studied. Similar results were reported by Syed *et al*., 2019; Pooja *et al.,* 2020 [29] and [30]. Some of the gel images with 29 genotypes showing polymorphism were shown in Fig. 2 (Fig. 2a to Fig. 2c).

3.3.2 Dendogram analysis

By using UPGMA (Unweighted Pair Group Method with Arithmetic Mean), cluster analysis was done to determine the phylogenetic relationship among the 29 rice genotypes. A dendogram (Fig. 3) was constructed based on Jackard's dissimilarity coefficient indicating the 29 genotypes to be grouped into 2 main clusters *i.e*. cluster I and cluster II with dissimilarity coefficient of 0.34. Cluster I was further sub-divided into two minor sub-groups IA and IB with dissimilarity coefficient 0.57. Cluster

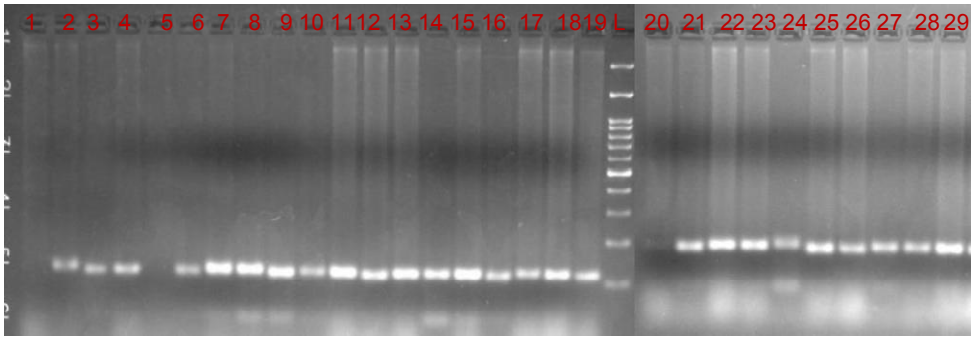

Fig. 2a. Gel image showing banding profile obtained by RM 227

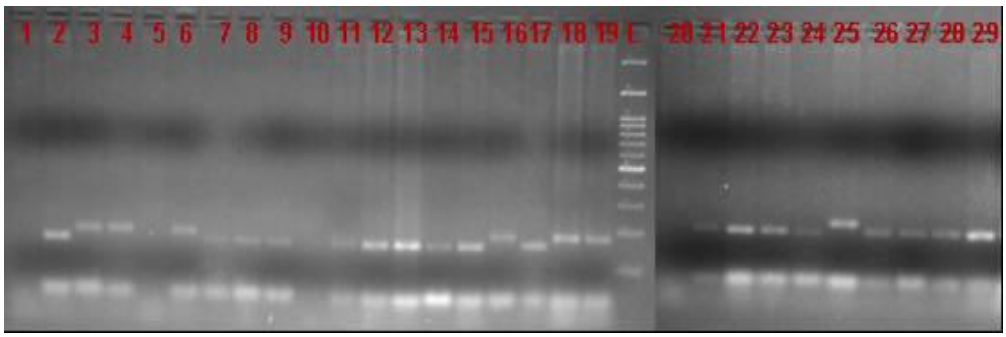

Fig. 2b. Gel image showing banding profile obtained by RM 11

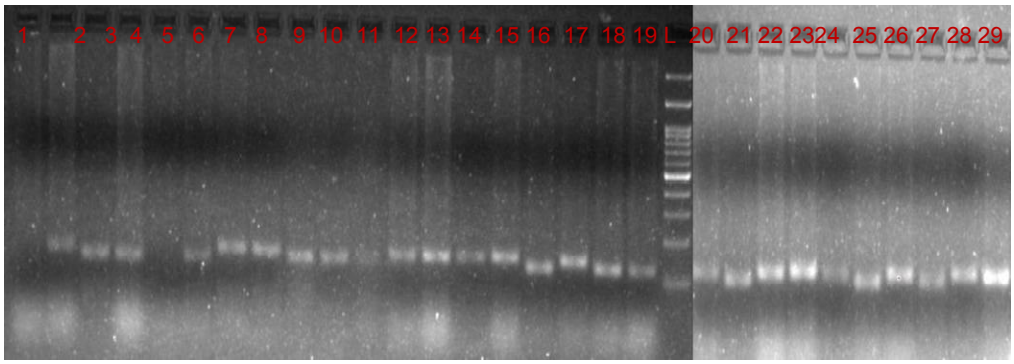

Fig. 2c. Gel image showing banding profile obtained by RM 259

Fig. 2. Gel images of 29 local rice genotypes analysed using SSR markers. Lane 1-29 represented the rice genotypes as listed in Table 1 and L is the Ladder (Number 19, 21 and 22 are check genotypes)

IA was further sub-divided into 2 subgroups *i.e.* IA-1 and IA-2 (0.59). Cluster IA-1 contains 3 genotypes and cluster IA-2 contains 6 genotypes. Cluster IB contains 2 genotypes. Cluster II was also further sub-divided into 2 minor sub-groups *i.e.* II-A and II-B with dissimilarity coefficient of 0.41. Cluster II-A was further sub-divided into 2 subgroups *i.e.* IIA-1 and IIB-2 (0.54). Cluster IIA-1 contains 6 genotypes and cluster IIB-2 has 4 genotypes. Cluster II-B contains 8 genotypes. The genotypes included in different clusters were mentioned in Table 5. Therefore, for utilization in breeding programmes it is very essential to select desirable genotypes from the most diverse clusters with higher dissimilarity coefficient.

3.3.3 Jackard dissimilarity coefficient

Among the genotypes studied the dissimilarity coefficient was used to determine the level of relatedness. The dissimilarity coefficient varies from 1 to 0, the value which is more close to 0, shows high similarity while those values which are close to 1 shows high dissimilarity. The dissimilarity coefficient ranged from 0.6373 to 0.7322. The total average of dissimilarity coefficient of all 29 rice genotypes is 0.68. The dissimilarity coefficient value was largest between the cultivar Kalanamak-2 and HUR-1304 (0.851); cultivar Anupam gold and HUR-1309 (0.851) followed by cultivar BHULC-8 and HUR-1309 (0.833). The lowest value was found between the cultivar BHULC-8 and Anupam Gold (0.421) followed by Vishnu Bhog white and Pusa Basmati 1121 (0.475) and G.R. 32 and BHULC-56 (0.478). Similar results were found by [29,30,31,32]. Hence, according to the dendogram obtained (Fig. 3) and Jackard dissimilarity coefficient, most diverse cultivars are Kalanamak-2 and HUR-1304; Anupam Gold and HUR -1309, followed by BHULC-8 and HUR-1309 whose results are a bit similar with the *inter se* genetic distances obtained from the morphological diversity analysis.

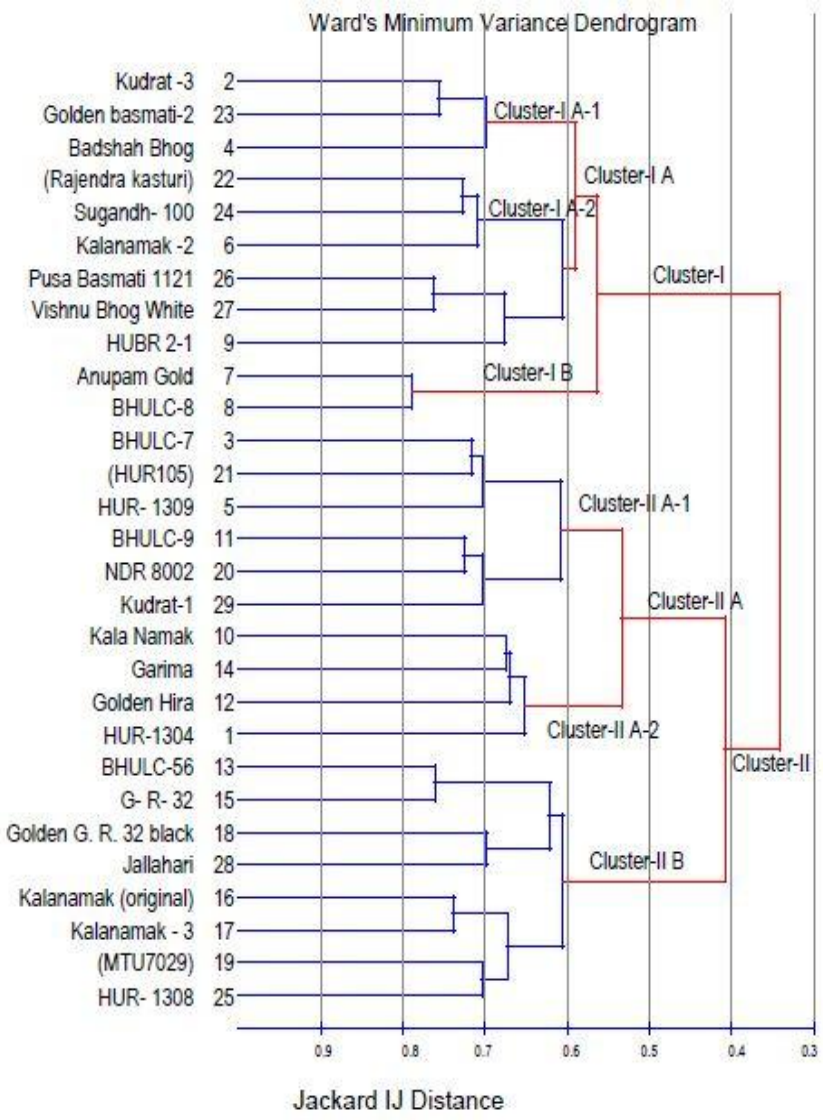

Fig. 3. Dendrogram dividing the genotypes into clusters based on Jackard IJ distance (MTU-7029, HUR 105 and Rajendra kasturi are the check genotypes

Table 5. Grouping of 29 rice genotypes into different clusters based on Jackard IJ coefficient

S. no.	Cluster name	Number of genotypes	Name of the genotypes
1.	IA-1	3	Kudrat-3, Golden basmati-2, Badshah Bhog
2.	IA-2	6	Rajendra Kasturi, Sugandh-100, Kalanamak-2, Pusa Basamati 1121, Vishnu Bhog white, HUBR 2-1
3.	IB	2	Anupam Gold, BHULC-8
4.	IIA-1	6	BHULC-7, HUR-105 ,HUR-1309, BHULC-9, NDR 8002, Kudrat-1
5.	IIA-2	4	Kalanamak, Garima, Golden Hira, HUR-1304
6.	IIB	8	BHULC-56, G.R.-32, Golden G.R.32, back Jallahari, Kalanamak (original), Kalanamak-3, MTU7029, HUR-1308

4. CONCLUSION

Wide range of variability was exhibited by the genotypes for most of the traits. Analysis of variance showed a considerable difference among the genotypes for all the traits under study. The genotype, MTU7029 (one of the checks) was found to be the best for yield and yield traits on the basis of per se performance. However, the above study of diversity concluded most diverse as Anupam gold and HUR-1309; Kalanamak-2 and HUR-1304 followed by BHULC-8 and HUR-1309. In the view of crop improvement, breeders may attempt hybridization among the above genotypes which showed maximum diversity, for creating more genetic variability in rice and to help in development of promising rice cultivars. It has to be kept in mind that both morphological and molecular techniques to study diversity present in the germplasm have been proven useful and one technique complements the other and provides a reliable result.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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