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RESEARCH PAPER

Assessment of Genetic Variation in the Indigenous Rice Cultivars (*Oryza sativa* L.) of Bangladesh

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ABSTRACT

Rice belongs to the family Gramineae which possesses enormous genetic diversity. A total of twenty six rice cultivars of Bangladesh were utilized in this study to investigate the genetic variation and molecular genotyping by using five microsatellites markers *viz,* RM25, RM251, RM259, RM307, RM413. Observed heterozygosity (Ho), expected heterozygosity (H*e*), number of effective alleles (N*e*), and polymorphism information contents (PIC) were calculated. The heterozygote deficiency or excess was detected by the fixation index (F_{is}). A total of 59 alleles were detected from five selected SSR primers. Results indicated that the number of alleles per locus ranged from 10 to 13. The genetic distance for all possible pairs of rice variety combination ranged from 0.000 to 2.890. The values of polymorphic information content (PIC) varied from 0.8484 to 0.9124, and according to the result the primer RM307 was found to be the most polymorphic. The dendrogram prepared on the basis of similarity matrix using the UPGMA algorithm based on Nei's (1972) genetic distance delineated the used rice varieties into four major clusters with several sub-clusters. This molecular genotyping approaches will be helpful for exploiting varietal identification and conservation of rice cultivars in future.

Key words: Genetic variation, indigenous cultivars, rice, Bangladesh

Introduction

Rice, *Oryza sativa* (2n=24) is one of the oldest domesticated crop species in the world which is also considered as the predominant staple food and a major source of dietary carbohydrate or energy for more than half of the world's population (Wennberg 2014). Bangladesh is mainly a rice growing country. The contribution of the crop sector to gross domestic product (GDP) is 10.98% while rice alone contributes 12.5 percent of total agricultural GDP (BBS, 2018).
Genetic variation in rice has been studied by various means such as biometrical analysis, biochemical analysis (isozyme) and molecular markers (Chakravarthi and Naravaneni 2006). Polymerase chain reaction (PCR) based and DNA sequence based molecular markers are the major means of molecular genetic diversity study which are commonly being exploited (Gupta *et al.* 1999; Jones *et al.* 1999; Winter and Kahl 1995). The most widely used DNA markers include Single Nucleotide Polymorphism (SNP), Simple Sequence Repeats (SSR), Restriction Fragment Length Polymorphism (RFLP), Random Amplification of Polymorphic DNA (RAPD) (Williams *et al.* 1990; Karp *et al.* 1996) and Amplified Fragment Length Polymorphism (AFLP) (Vos *et al.* 1995). Molecular markers, in general, have proven to be very useful for crop improvement and studies of crop evolution in many species (Mohan *et al.* 1997). Due to their abundance, DNA or molecular markers have been used extensively to assess the level of genetic diversity in most crop species (Islam *et al.* 2007). SSRs are also known as microsatellite molecular markers based on tandem repeats of short (2-6bp) DNA sequences (Litt and Lutty 1989). These DNA sequences are highly polymorphic even among closely related cultivars due to mutation causing variation in the number of repeating units (Saghai-Maroof *et al.* 1994). Microsatellite markers are reported to be more variable than RFLPs or RAPDs thus have been widely adopted for genetic studies in humans (Dib *et al.* 1996) and other mammals (Sun and Kirkpatric 1996). It can be analyzed by a rapid, technically simple and inexpensive PCR based assay that requires only small quantities of DNA. Through PCR reactions, different alleles at a locus can be detected by using conserved DNA sequences flanking the microsatellite primers. Microsatellite markers are co-dominant and can be transmitted in simple Mendelian segregation. Lastly, microsatellites are abundant and uniformly distributed in plant genomes (Lagercrantz *et*

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al. 1993; Wang *et al.* 1994; Akkaya *et al.* 1995). Microsatellite markers are utilized in plant breeding programs, particularly for the identification of accessions, detection of genetic relationships within and among genotype, measurement of the structure and quantity of genetic variation and for the identification and localization of particular genes or DNA sequences (Temnykh *et al.* 2000; 2001). Hence, the sequence data of various crops have revealed for conservation among the genomes through the regions flanking by the microsatellite loci.

In Bangladesh, 20 crop species including rice, wheat, maize, barley, rapeseed, soybean, potato and other crops were tested with SSR by Rahman and his associates (2006; 2007; 2008; 2009; 2010; 2014) to know the phenotypic and genotypic variation with a view of documentation and characterization. The current study was continuity to the above research, and designed for detecting genetic diversity, genetic variation in indigenous rice cultivars grown in Bangladesh for further development of commercial rice varieties.

Materials and Methods

Raising of Seedlings

All the selected rice cultivars were grown in the net house on the roof of the Dept. of Genetics & Plant Breeding, Bangladesh Agricultural University. It was maintained in plastic pots in soil mixed with required amount of compost and fertilizers for each and individual pot. Three replications with two plants in each pot were allowed to grow.

Extraction of Plant Genomic DNA

Total genomic DNA was extracted from the fresh and juvenile leaf samples of 22-days-old seedling of each 26 rice cultivars following the protocol described by Aljanabi and Martinez (1997) and also used by Rahman *et al.* (2007; 2008; 2009; 2010) with some modifications (Modified CTAB method). DNA sample of each cultivar after dissolving in 30-40µl of TE buffer was preserved separately at -20°C.

Quantification and Optimization of DNA Concentration

Existence of genomic DNA was confirmed on 1% agarose gel qualitatively. The amount of genomic DNA was quantified at 260nm spectrophotometrically (Spectronic® Genesis™). Using the absorbance reading obtained for DNA sample of each rice variety, the original DNA concentrations were determined and adjusted to 25 ng/µl finally.

Assortment of Microsatellite Primers

A set of twenty one microsatellite primer pairs (RM5, RM25, RM55, RM105, RM151, RM153, RM170, RM206, RM251, RM259, RM264, RM266, RM278, RM287, RM307, RM333, RM334, RM335, RM413 RM475 and RM481) distributed in the rice genome were identified from the available data-base (http://www.gramene.org/) for rice SSR markers as described by Akagi *et al.* (1996), Panaud *et al.* (1996), Temnykh *et al.* (2000; 2001) and McCouch *et al.* (2002). From those identified primers, at first 3 to 5 primers were tested against 5 randomly selected cultivars with a recommended PCR thermal profile. Primers were selected on the basis of band resolution intensity, presence of smearing, consistency within individuals and potential for population discrimination. In this way, five primer pairs *viz*., RM25, RM251, RM259, RM307 and RM413 representing chromosome numbers 8, 3, 1, 4 and 5 of rice genome (Temnykh *et al.* 2001) with clear and expected amplified product sizes were selected and used for microsatellite analysis in this study. However, the selected primers were then screened against all the 26 varieties at once.

PCR amplification

The amplification conditions were based on Panaud *et al.* (Panaud *et al.* 1996; Temnykh *et al.* 2000; 2001) with minor modifications suggested by previous research (Alam *et al.* 2016; Rahman *et al.* 2006; 2007; 2008; 2009; 2010). Eppendorf™ oil-free thermal cycler gradient was utilized in this PCR reaction. Approximate 15 µl reaction volume consists of 50ng sample DNA (2.0 µl), 10x PCR Buffer (3.0 µl), 1 µM of each forward and reverse SSR primer (1.0 µl), 0.25 mM dNTPs (1.5 µl), 1 unit ampli Taq DNA polymerase (0.5 µl), and nuclease free double distilled deionized water (6.0 µl) were utilized to perform PCR reaction. PCR settings were carried out by the conditions described by the previous research (Alam *et al.* 2016; Rahman *et al.* 2006; 2007; 2008; 2009; 2010) as follows: 95°C for 5 min (an initial denaturation) followed by entire 35 cycles, 95°C for 40 sec (denaturation), 55°C for 30 sec (annealing) and 72°C for 1 min (elongation/extension), then a final elongation/extension cycle at 72°C for 7 min. Amplified PCR reactions were then stored at –20°C for further utilization.

Agarose Gel Electrophoresis and Documentation

Top vision submarine horizontal electrophoresis system (BIORAD™ Sequencing Cell) was utilized to electrophoresis the PCR reactions (Figure 1). Prior to electrophoresis, each 07 µL amplified PCR aliquot and 3 µL of loading buffer (Rahman *et al.* 2010) were mixed gently. This mixture was then loaded on 2% agarose gel, and placed into the submergible horizontal gel chamber with 1x TBE running buffer (Trizma base, boric acid and EDTA; p^H 8.0). A five microlitre (5 µL) 100 bp standard DNA (Gene ruler, Fermentas®) ladder was added in both left and right side of the gel to compare the molecular weight of the amplified PCR products of each cultivar. PCR mixtures (10 µl) were subjected to electrophoresis at 100V and 50W for 2 hrs 40 mins. The electrophoresis sample was then kept as photographic image by the camera Polaroid gel documentation system (UVP, BioDoc-It™ System).

Genotyping and Microsatellite Data Analysis

All distinct unambiguous DNA fragments/bands representing particular alleles (A, B, C, etc.) at the microsatellite loci were scored visually and given identification numbers according to their heading position on gel. DNA FRAG version 3.03 computer software was used to estimate allelic length (Nash 1991). The allele frequency data (DNA fragment) was exported as diploid datasheet arrangement (AA, AB, CC and so on) on POPGENE v1.31 computer program (Yeh *et al.* 1999) , and therefore, utilized for the several statistical analysis including “observed number of alleles-N*a* (Saavedra and Harrier 2001)”, “effective number of alleles-N*e*” (Kimura and Crow 1964), “allelic diversity index (PIC=1-$\sum X_i^2$, where X_i indicates the frequency

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of the ith allele), Shannon's Information index-*I* (Lewontin 2014)", "Hardy-Weinberg equilibrium (H*e* and H*o* of Levene 1949) and Gene flow-N_m)", "Nei's gene diversity index (*h*) (Nei 1972; Nei 2013)", "chi-square & probability index", and "Wright's fixation index-F_{is} (Nei 1977)". Finally, Nei's (1972) genetic distance (*D*) and similarity values (Table 4) were utilized to generate an UPGMA (Unweighted Pair Group Method of Arithmetic Means) phylogenetic tree (Figure 2) (Evening 2012) which was visualized through the "Treeview" computer software (Page 1996). The formatted several clusters on UPGMA dendrogram (Figure 2) were then exploited to clarify the interaction among the cultivars in this investigation.

Results and Discussions

Microsatellite has become one of the most widely used molecular markers for genetic studies in recent years. The specific adaptor-amplified DNA fragments of this analysis is a very simple and efficient approach that can be successfully applied for a number of plant genomes.

Allelic Variation

All the amplified microsatellite loci (RM25, RM251, RM259, RM307 and RM413) were found to be polymorphic. The microsatellite profiles of loci RM25, RM251, RM259, RM307 and RM413 are shown in Figure 1. Using five primers across 26 varieties 59 allele were identified. The number of alleles ranged from ten to thirteen per locus. The locus RM307 and RM413 had the highest number of alleles (13) while the locus RM25, RM251 had the lowest number of alleles (10, 11) and RM259 had twelve alleles (Table 1). The effective number of allele was the highest for RM413 (10.24) and the lowest for RM259 (6.28) shown in Table 2. Yang *et al*. (1994) found up to 25 alleles for 10 microsatellite markers among 238 accessions of *Indica* and *Japonica* cultivars and landraces.

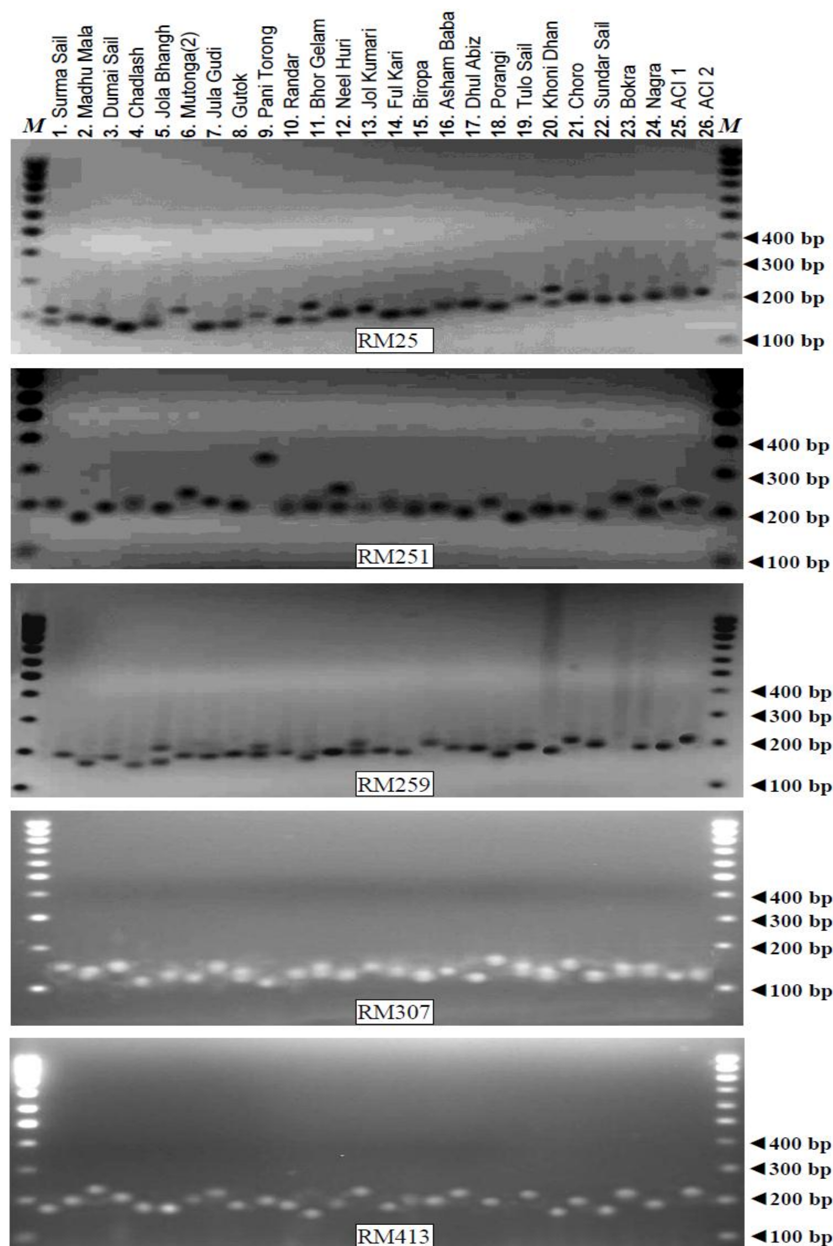

Figure 1: Microsatellite profile of five SSR markers *viz.,* RM25, RM251, RM259, RM307 and RM413by using 26 indigenous rice cultivars grown in Bangladesh, M: 100bp DNA ladder

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Table 1: Size and frequency of alleles and diversity index at 5 SSR loci across 26 rice *(Oryza sativa* L.) varieties

Allele	**RM25** (Chromosome 8)	**RM251** (Chromosome 3)	**RM259** (Chromosome 1)	**RM307** (Chromosome 4)	**RM413** (Chromosome 5)
Allele A	0.0577	0.0577	0.0385	0.0577	0.0192
Allele B	0.0962	0.0962	0.0192	0.0192	0.0577
Allele C	0.1154	0.0577	0.0192	0.0385	0.1154
Allele D	0.1923	0.1731	0.0385	0.1731	0.1154
Allele E	0.1154	0.0577	0.0385	0.0577	0.0769
Allele F	0.0577	0.2115	0.0385	0.0192	0.0769
Allele G	0.2308	0.0769	0.1154	0.1538	0.0769
Allele H	0.0192	0.1923	0.2308	0.0769	0.1731
Allele I	0.0769	0.0385	0.2692	0.1154	0.0769
Allele J	0.0385	0.0192	0.0769	0.0769	0.0962
Allele K		0.0192	0.0769	0.0769	0.0385
Allele L			0.0385	0.0769	0.0385
Allele M				0.0573	0.0385
Diversity index (PIC=1-$\sum Xi^2$)	0.8594	0.8613	0.8484	0.9124	0.9068
Observed number of alleles	10	11	12	13	13
Sequence of primers (5´-3´)	**F:***ggaaagaatgatcttttcatgg* R:*ctaccatcaaaaccaatgttc*	**F:***gaatggcaatggcgctag* **R:***atgcggttcaagattcgatc*	**F:***tggagtttgagaggaggg* **R:***cttgttgcatggtgccatgt*	**F:***gtactaccgacctaccgttca* **R:***ctgctatgcatgaactgctc*	**F:***ggcgattcttggatgaaga* **R:***accccaccaaacaagac*
Motif	***(ga)***$_{18}$	***(ct)***$_{29}$	***(ct)***$_{17}$	***(at)***$_{14}$***(gt)***$_{21}$	***(ag)***$_{11}$

Table 2: Summary of genetic variation and heterozygosity statistics for all loci

Locus	N*a**	N*e**	*I**	F_{st}	Nm*	Obs_ Hom	Obs_ Het	Exp_ Hom*	Exp_ Het*	*h**	Ave_ Het
RM25	10.00	7.11	2.11	0.955	0.011	0.923	0.076	0.123	0.876	0.859	0.038
RM251	11.00	7.19	2.14	0.732	0.091	0.538	0.461	0.122	0.877	0.860	0.230
RM259	12.00	6.28	2.11	0.954	0.012	0.923	0.076	0.142	0.857	0.841	0.038
RM307	13.00	9.72	2.40	0.935	0.017	0.884	0.115	0.085	0.914	0.897	0.057
RM413	13.00	10.24	2.43	0.914	0.023	0.846	0.153	0.079	0.920	0.902	0.076
Mean	11.80	8.11	2.24	0.894	0.031	0.823	0.176	0.110	0.889	0.872	0.088

*N***a** = Number of observed alleles
*N***e** = Number of effective alleles =$1/(\sum X_i^2)$, Where X_i is the frequency of the ith allele (Kimura and Crow 1964)
****I*** = Shannon's Information index by Lewontin (2014)
*Unbiased Expected Homozygosity and Heterozygosity were computed using Levene (1949)
*$\mathbf{N_m}$ = Gene flow estimated from $F_{st} = 0.25(1 - F_{st})/F_{st}$.
****h*** = Nei's gene diversity index (Nei 1972; Nei 2013)

Genetic Variation

Genetic differentiation (F_{st}) values were recorded in the ranges from 0.732 to 0.955 with an average of 0.894 and gene flow (N*m*) values varied from 0.011 to 0.091 with an average of 0.031 (Table 2) indicating comparatively higher level of genetic differentiation and low level of gene flow in 26 rice (*O. sativa* L.) cultivars which were expression of diversity among the varieties studied (Sharma *et al.* 2016). Variation also found in number of alleles, observed and expected heterozygosity (H*o* and H*e*). Across 26 rice cultivars, RM251 showed highest average heterozygosity (0.230) while RM25 and RM259 (0.035) yielded lowest average heterozygosity. Over all Nei's genetic diversity (*h*) ranged from 0.841 to 0.902. Variation also found in allele frequency and diversity index values where diversity index (PIC) ranged from 0.848 to 0.912. The highest diversity index was recorded for RM307 marker (0.912). Lower PIC value indicates the result of closely related genotypes and higher PIC values might be the result of diverse genotypes (Temnykh *et al.* 2000; 2001, Khalequzzaman *et al.* 2017). A wide range of allele frequency along with PIC value, major genetic variation in sense of observed and expected heterozygosity (H*o* and H*e*) were detected in this investigation. Similar observations were recorded in several Bangladeshi local rice cultivars by various previous researchers (Siddique *et al.* 2016; Rahman *et al.* 2006; 2007; 2008; 2009; 2010; 2014). These values have suggestive indication for the diversity analysis

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of several geographical indicating populations (Sharma *et al.* 2016).

The ranges of wrights fixation index {F_{is} =(H*e* - H*o*) / H*e*} were recorded 0.0182-1.00; 0.0566-1.00; 0.0182-1.00; 0.6478-1.00 and 0.4615-1.00 to the five primers RM25, RM251, RM259, RM307 and RM413, respectively (Table 3). It is usually the parameters for the measurement of heterozygote deficiency or excess for the selected molecular markers. However, the values of F_{is} (Table 3) in this investigation were the indication of the presence of high genetic diversity among the cultivars (Jasim *et al.* 2018), thus it can be considered for the selection of these cultivars without QTLs research (Chen *et al.* 2017).

Table 3: Wright's (1978) fixation index (F_{is})

Allele \ Locus	RM25	RM251	RM259	RM307	RM413
Allele A	0.018	0.018	1.000	0.708	1.000
Allele B	1.000	0.018	1.000	1.000	0.780
Allele C	0.780	0.787	0.647	1.000	0.836
Allele D	1.000	0.056	1.000	1.000	0.660
Allele E	0.923	0.899	0.018	1.000	1.000
Allele F	1.000	1.000	1.000	1.000	0.461
Allele G	1.000	0.647	0.647	1.000	0.886
Allele H	1.000	0.461	1.000	0.647	0.780
Allele I	1.000	0.647	1.000	1.000	1.000
Allele J	0.018	0.461	0.886	1.000	0.647
Allele K	1.000	0.037	1.000	1.000	1.000
Allele L			0.780	0.780	
Allele M			0.836	0.647	
Allele N			1.000	1.000	
Allele O				0.780	

*F_{is} = Wright's fixation index (F_{is}) as a measure of heterozygote deficiency or excess (H*e* - H*o*) / H*e*) (Nei, 1977)

Genetic Distance (*D*) and Cluster Analysis

Geographical distance is an important factor that influences the genetic relatedness of populations (Wright 1978). Among the 26 rice cultivars, a total of 325 varietal pairs are achievable. However, the summary of Nei's genetic distance (*D*) from 325 varietal pairs among 26 rice cultivars varied from zero to 2.890 (Table 4). Overall, 22 varietal pairs (6.77%) exhibited the highest Nei's genetic distance (2.890) *viz,* Porangi× Chadlash, Choro × Chadlash, Choro × Porangi etc.; the lowest genetic distance (0.00) was calculated in 129 combinations (39.69%) such as Madhu Mala × Surma Sail, Chadlash× Surma Sail, Jola Bhangh × Madhu Mala etc. (Table 4). The rest 174 (53.54) varietal pairs exhibited fairly constructive genetic distance among them. This intimacy may be feasible in the genetic make-up of the locus for which the primers were responsible to distinguish along with low ecotype variation (Rahman *et al.* 2006; 2007; 2008; 2009; 2010; 2014). In addition, UPGMA dendogram based on Nei's (1972) genetic distance indicated segregation of 26 varieties of rice into four major clusters (I to IV). Cluster I consisted of thirteen cultivars, while clusters II and III abided different ten and two cultivars, respectively. Cluster IV reposed the lowest number of accessions with single rice cultivars (Choro) that differed from other clusters containing rest 25 varieties into three major groups (Figure 2). The spot in between 2.890 to 2.249 genetic distances on figure 2 flicked its primary cluster separation. Consequently, moving to the null genetic distance, rest clusters were divided into proportionate sub-clusters with particular genetic distance and similarity. However, the clusters were separated into sub-cluster, and sub-clusters subsequently further separated into themselves as remarking A, B, C, D, E, F and so on (Figure 2). The dendrogram showed that the varieties were closely related belonging to the same cluster while the cultivars choro, biropa and bhor gelam belonging to different cluster on Figure 2 suggesting that these varieties were genetically diverse in origin. Hence, it can provide ample information to seek out the best-performing cultivars for further breeding programs in upcoming future.

Table 4: Summary of Nei's (1972) genetic distance (*D*) among 26 rice varieties

Varieties	Surma Sail	Madhu Mala	Dumai Sail	Chadlash	Jola Bhangh	Mutonga(2)	Jula Gudi	Gutok	Pani Torong	Randar	Bhor Gelam	Neel Huri	Jol Kumari	Ful Kari	Biropa	Asham Baba	Dhul Abiz	Porangi	Tulo Sail	Khoni Dhan	Choro	Sundar Sail	Bokra	Nagra	ACI 1	ACI 2
Surma Sail	****																									
Madhu Mala	0.000	****																								
Dumai Sail	2.191	0.752	****																							
Chadlash	0.000	0.000	0.000	****																						
Jola Bhangh	1.609	0.000	0.805	0.000	****																					
Mutonga(2)	0.000	1.674	1.498	0.000	0.916	****																				
Jula Gudi	1.609	0.000	1.498	0.000	0.000	0.000	****																			
Gutok	2.191	1.498	0.000	0.752	1.674	0.000	2.191	****																		
Pani Torong	0.000	1.504	0.000	1.504	0.000	1.674	2.250	2.832	****																	
Randar	0.000	1.557	0.805	0.000	0.916	0.000	1.609	2.191	2.890	****																
Bhor Gelam	1.498	2.890	2.079	1.445	2.890	1.092	2.191	1.163	2.832	1.498	****															
Neel Huri	0.000	0.000	2.079	0.000	2.191	0.000	0.582	2.079	1.040	1.092	2.890	****														
Jol Kumari	0.000	1.504	0.000	1.504	0.000	2.250	0.000	1.445	0.811	0.000	1.222	2.138	****													
Ful Kari	0.916	2.250	2.191	0.000	0.916	1.609	1.609	1.674	1.557	0.000	2.890	1.498	2.250	****												
Biropa	0.000	2.890	1.498	2.250	2.890	0.000	0.000	2.191	1.557	1.609	0.000	2.890	2.250	2.890	****											
Asham Baba	0.000	2.250	0.000	1.557	0.000	0.000	1.609	1.498	0.000	0.000	2.890	1.498	1.557	1.609	2.890	****										
Dhul Abiz	1.498	1.151	1.498	0.000	0.000	1.498	0.000	0.000	0.000	1.609	0.000	0.000	0.000	1.609	0.000	0.511	****									
Porangi	0.000	1.504	1.445	2.890	0.000	0.000	1.674	0.000	0.000	1.557	0.000	0.000	0.000	0.000	2.250	0.000	1.557	****								
Tulo Sail	2.890	2.832	0.000	2.138	2.890	0.000	0.000	1.092	2.638	0.000	0.000	2.890	1.498	2.191	0.000	2.191	2.191	2.832	****							
Khoni Dhan	0.000	2.832	1.163	0.000	0.582	1.092	0.000	0.000	1.674	1.498	1.386	2.079	0.000	1.092	1.498	2.191	2.191	1.040	2.773	****						
Choro	2.890	0.000	1.445	2.890	1.557	2.890	0.000	0.000	0.000	2.890	0.000	2.138	0.000	0.000	1.557	0.000	0.000	2.890	2.832	1.445	****					
Sundar Sail	0.000	0.000	1.092	0.000	0.000	1.498	1.092	1.674	0.000	0.000	2.191	0.000	1.092	1.674	0.000	1.498	1.609	0.000	1.498	0.000	0.000	****				
Bokra	0.000	1.498	2.832	1.674	1.151	1.151	0.000	0.000	1.674	0.000	1.445	0.000	0.000	1.557	1.557	0.000	0.000	2.890	1.222	1.222	2.890	0.000	****			
Nagra	1.498	1.092	1.674	0.000	1.498	2.191	0.000	0.000	0.000	2.191	0.000	2.773	1.498	0.000	1.092	1.092	1.674	2.832	2.773	1.674	0.886	1.498	1.040	****		
ACI 1	0.000	0.000	2.191	0.000	1.609	1.609	1.092	2.191	0.000	1.674	0.000	0.000	0.000	0.000	1.609	0.000	0.000	0.000	2.805	2.191	0.000	0.000	0.641	1.092	****	
ACI 2	1.498	0.000	0.000	1.445	0.000	0.000	1.498	0.981	2.138	1.498	0.981	1.386	1.445	0.000	0.000	1.498	0.000	2.138	0.000	2.773	1.040	2.191	0.000	0.886	0.000	****

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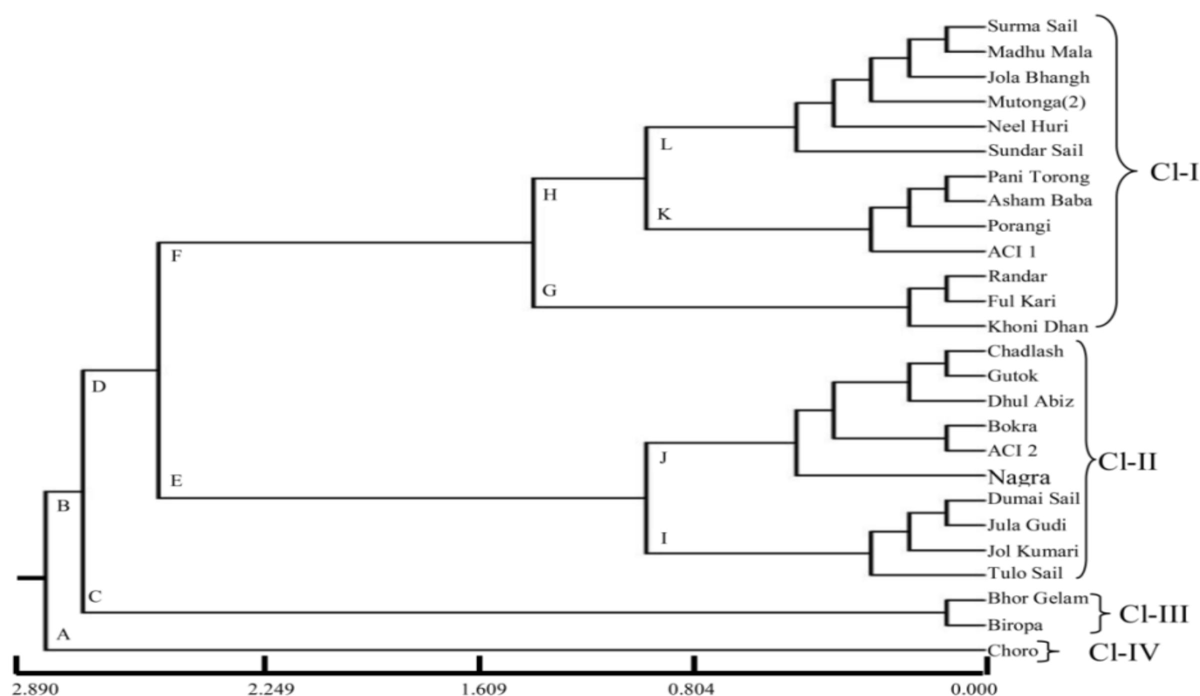

Figure 2: UPGMA dendrogram based on Nei's (1972) genetic distance, summarizing the data on differentiation among 26 rice varieties according to microsatellite marker analysis

Conclusion

Microsatellite markers showed genetic variability in the studied indigenous rice cultivars grown in Bangladesh and they are powerful tools for estimating genetic similarities and diversity. The genetic relationships presented among the cultivars are helpful for future breeding programs through selection of genetically diverse parents. The present work was the continuity of the previous study to characterize and detect genetic variation of rice varieties of Bangladesh. The results indicated that the present study might be used as a guideline for developing mapping population, marker assisted selection (MAS) and crop improvement of rice varieties, and consequently enables a genetic conservation plan in Bangladesh.

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