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Genetic Diversity Analysis in Boro Rice *(Oryza sativa* L.*)* Landraces of Bangladesh Using SSR Markers

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ABSTRACT

Genetic diversity and associations among 96 Boro rice landraces of Bangladesh were assessed using 12 SSR markers at the Genetic Resources and Seed Division of Bangladesh Rice Research Institute (BRRI). The number of alleles per locus ranged from 11 (RM163) to 41 (RM283), with a mean of 21 alleles. Polymorphism information content (PIC) ranged from 0.75 (RM163) to 0.95 (RM283) with an average of 0.891, showing a large variability among the tested landraces. PIC values showed that RM283 was considered as the best marker for identifying and evaluating the diversity of Boro rice landraces. The frequency of most common alleles at each locus ranged from 8% (RM283) to 34% (RM275, RM277). Nei's genetic distance based UPGMA dendrogram grouped the studied landraces into seven different clusters with a similarity coefficient of 0.11. Based on the dendrogram constructed using SSR markers, the accessions that are distant from each other in terms of genetic distance and diversity index (such as Pashusail and Tulsi Boro; Raja sail and Kali Boro; Bashful and Jamir; Begun bitchi and Boro deshi; Banjira and Bogra (Deshi); Jagli Boro and Lahi Boro; Bimion and Gorchi sail; Jhati sail and Khaia Boro; Tepi Boro and Jamir Boro) are strongly recommended for selection as parents in future breeding programmes aimed at developing high-yielding and stress-resilient rice variety in contribution to global food security. The results of this study could be useful for the identification of genetically diverse parents from the landraces for breeding programme aimed at varietal improvement.

Key words: Rice (*Oryza sativa* L.), boro, genetic distance, dendrogram, landrace, varietal improvement.

INTRODUCTION

Rice and food security are considered as the same thing in Bangladesh context (Brolley, 2015). Rice is an important staple food and accounts for about 78% of the country's total net cultivated area. The country achieves self-sufficiency to meet the rice demand of a population of 169.04 million with 11.55 million hectares of gross cultivated area (Kabir *et al.,* 2020; Nasim *et al.,* 2021). Bangladesh ranks fourth in the world in rice consumption, with an annual per capita availability of about 213.5 kg (FPMU Database, 2020). Current rice consumption is approximately 328.9 g per capita per day, providing approximately 60% of total calories and 50% of total protein for adults (HIES, 2022). About 48% of rural workers participate directly or indirectly in rice production for their livelihood. Rice is grown all year round in three seasons: Aus, Aman and Boro. Since independence, rice production has increased

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three-fold from approximately 11 MT in 1971–72 to about 36.6 MT in 2019–20 (BBS, 2020). The contribution of rice to the value of the crop sub-sector is about 70% (Mottaleb and Mishra, 2016).

Due to the great importance of this crop and its close connection to food security and local lifestyle and culture, Asian farmers have been selecting and cultivating a wide range of rice varieties for thousands of years. It is estimated by scientists that more than 1,40,000 rice varieties have been developed/selected/isolated in Asia. More than 1,32,000 germplasm are preserved in the world's largest rice genebank, the International Rice Research Institute (IRRI), located in the Philippines (http://irri.org/our-workresearch/genetic-diversity). A total of 9,006 varieties/landraces/cultivars/wild genotypes has been collected from indigenous and exotic sources and preserved in the BRRI Genebank. Genetic diversity is the primary component of any agricultural production system for selection, conservation, characterization, and appropriate use of germplasms (Emon and Ahammed, 2020). The lack of genetic diversity limits crop production and represents a serious problem in any breeding programme aimed at varietal development. Therefore, studying natural diversity or creating genetic diversity (if not exist) is a crucial step to increase yield. Rice breeders are using different types of germplasm like landraces, wild stock, commercially approved varieties etc. to explore the genetic diversity within the existing germplasm to increase yields. However, studies on the genetic diversity of different varieties and germplasm of Boro rice in Bangladesh are rare. Information about genetic diversity and the relationship among available germplasm is essential before taking any rational plant breeding programme. The genetic diversity of crop species can be determined using morphological, biochemical and molecular (DNA) markers (Rao, 2004). Several researchers have used morphological traits for diversity assessment and characterization of Bangladesh rice germplasm (Siddique *et al.,* 2011; Banik *et al.,* 2012; Khalequzzaman *et al*., 2012; Baktiar *et al.,* 2013; Siddique *et al.,* 2013; Islam *et al.,* 2014; Ahmed *et al.,* 2015a, 2015b; Kulsum *et al.,* 2015; Akter *et al.,* 2016; Biswash *et al.,* 2016; Siddique *et al.,* 2016a; Akter *et al.,* 2017; Islam *et al*., 2017; Akter *et al.,* 2018; Islam *et al.,* 2018a; Siddique *et al.,* 2018; Islam *et al.,* 2019; Muti *et al*., 2020; Khalequzzaman *et al.,* 2022a; Khalequzzaman *et al.,* 2023). However, compared to biochemical markers, morphological markers are strongly affected by environmental factors and are therefore dependent on the environmental conditions during cultivation. The limitations associated with morphological and biochemical markers are overcome by molecular markers (Rao, 2004). Molecular characterization through the use of molecular markers is independent of environmental influences. This characterization can be performed with plant DNA from any growth stages (Tatikonda *et al.,* 2009). Among the various molecular markers, polymerase chain reaction (PCR)-based simple sequence-repeat (SSR) markers (Gianfranceschi *et al.,* 1998) have become popular in analyses of genetic diversity.

Molecular markers have been used successfully in registration activities such as cultivar and variety identification (Mailer *et al.,* 1994; Bligh *et al.,* 1999). An effective technique for DNA fingerprinting is the efficient PCR amplification of tandem repeat sequences that have long been known to be polymorphic and widespread in plant genomes, called simple sequence repeats (SSRs) or microsatellite polymorphisms (Cregan, 1992; Morgante and Olivieri, 1993). Motif mutations of and flanking sequences as well as the distribution of microsatellites in a species' genome are used to reveal genetic variation and cultivar identity. In plants, SSRs have been shown to be highly informative and site-specific markers in many species (Akkaya *et al.,* 1992; Legarcrantz *et al.,* 1993; Wu and Tanksley, 1993, Rahman *et al.,* 2007). SSRs are becoming increasingly useful for integrating genetic, physical and sequence-based maps in rice, while providing breeders and geneticists with a powerful tool for linking phenotypic and genotypic variation.

Microsatellites are abundant and well distributed throughout the rice genome (Akagi *et al.,* 1996; McCouch *et al.,* 1997; Wu and Tanksley, 1993). SSRs are codominant, recognize a high level of allelic diversity, and are efficiently characterized by PCR (McCouch *et al.,* 2002). Several researchers used molecular markers for diversity assessment and DNA fingerprinting of Bangladesh rice germplasm (Rahman *et al.,* 2006; Rahman *et al.,* 2007; Rahman *et al.,* 2008; Rahman *et al.,* 2010; Siddique *et al.,* 2014; Siddique *et al.,* 2016b, 2016c, 2016d; Khalequzzaman *et al.,* 2017; Siddique *et al.,* 2017; Islam *et al.,* 2018b; Islam *et al.,* 2021; Akter *et al.,* 2022; Khalequzzaman *et al.,* 2022b; Saha *et al.,* 2022; Khalequzzaman *et al.,* 2023). This study was undertaken with 96 Boro rice landraces and used molecular traits for characterization and diversity analysis. The objectives of this study were to evaluate the genetic diversity assessment of 96 Boro rice landraces and determine the genetic relationships exist among these landraces at molecular level.

MATERIALS AND METHODS

Plant materials

Ninety-six Boro rice landraces of Bangladesh were tested (Table 1). Five-gram seeds of each entry were first germinated and then sown into earthen pots for DNA extraction.

Table 1. Rice landraces used in this study with their provenance.

Landrace	BRRI accession no.	Place of collection	Variety	BRRI accession no.	Place of collection
Banajira	7	Barishal	Lafai	1969	Kishoreganj
Pashusail	54	Habiganj	Sail Boro	1970	Kishoreganj
Cunail	178	Tangail	Gochi	1971	Kishoreganj
Bhaturi	179	Tangail	Biron	1972	Kishoreganj
Dholi Boro	180	Tangail	Bogura	2251	Kishoreganj
Grugu Boro	182	Tangail	Lahaya	2252	Kishoreganj
Boro	253	Mymensingh	Chhola Boro	2258	Kishoreganj
Ausha Boro	254	Mymensingh	Kolisha Boro	2260	Kishoreganj
Jagli Boro	255	Mymensingh	Goa Bish	2261	Kishoreganj
Tepi Boro	258	Mymensingh	Madhab Sail	2264	Kishoreganj
Kali Boro	260	Mymensingh	Mogol Sail	2266	Kishoreganj
Kaiaka Boro	262	Mymensingh	Gola Tepi	2267	Kishoreganj
Poshu Sail	929	Sylhet	Lafa	2268	Kishoreganj
Gorchi Sail	932	Sylhet	Beun Bichi	3952	Netrakona

Landrace	BRRI accession no.	Place of collection	Variety	BRRI accession no.	Place of collection
Jhati Sail	933	Sylhet	Rata Boro	3959	Netrakona
Bimion	934	Sylhet	Kori Topa	3960	Kishoreganj
Soiler Peena	935	Sylhet	Lal Dengi	3962	Kishoreganj
Khaia Boro	936	Sylhet	Panpiag	3963	Kishoreganj
Boro Deshi	1405	Pabna	KN – 1B -361-1312-27- 1	3976	BRRI
Muktahar	1468	Dhaka	Choudhury Sail	3980	Sunamganj
Banajira	1470	Dhaka	Ashani	3981	Sunamganj
Bash ful	1471	Dhaka	Madanga	3982	Sunamganj
Am Boro	1472	Dhaka	Laitra Sail	3983	Sunamganj
Lahi Boro	1474	Dhaka	Fena Ful	3984	Sunamganj
Jamri Boro	1475	Dhaka	Kowla	3985	Sunamganj
Mukta har	1650	Dhaka	Madlai	3989	Habiganj
Bati Boro	1670	Dhaka	Lara	3991	Habiganj
Jamir	1706	Faridpur	Binni	3993	Habiganj
Kali Boro	1707	Faridpur	Lal Boro	3994	Habiganj
Bawoi	1708	Faridpur	Gachi Boro	3995	Habiganj
Khoea	1709	Faridpur	Bachi Boro	4000	Habiganj
Ulia	1711	Faridpur	Birain	4001	Habiganj
Solai	1713	Faridpur	Sona Rata	4002	Habiganj
Sada Boro (deshi)	1714	Faridpur	Naula topa	4003	Habiganj
Chaita Boro	1716	Khulna	Hunga Boro	4004	Habiganj
Isamoti	1790	Jashore	Bimon	4005	Habiganj
Jaista Boro	1792	Jashore	Gobi Sail	4006	Habiganj
Natel Boro	1793	Jashore	Nata Boro	4007	Habiganj
Guchi Boro	1796	Kishoreganj	Chaula Birain	4008	Habiganj
Lakhai	1800	Kishoreganj	Kala Birain	4009	Habiganj
Lakhai	1801	Kishoreganj	Badal Boro	4011	Sylhet
Bogra (Deshi)	1802	Kishoreganj	Jomir Sail	4012	Sylhet
Tupa	1811	Kishoreganj	Gopal Beri	4013	Habiganj
Begun Bitchi	1813	Kishoreganj	Muirol	4014	Habiganj
Pankaij	1817	Kishoreganj	Polash	4017	Habiganj
Bash Boro	1818	Kishoreganj	Gasbar	4202	Netrakona
Raja Sail	1819	Kishoreganj	Gumir Sail	4203	Netrakona
Tulsi Boro	1968	Kishoreganj	Jalda IRRI	4206	Brahmanbaria

SSR markers

Twelve SSR markers (Table 2) were used for the diversity analysis.

Table 2. List of the 12 simple sequence repeat (SSR) markers used in this study.

Locus name	Chr.	Repeat motif	Forward primer	Reverse primer
RM 6	2	(AG)16	GTCCCCTCCACCCAATTC	TCGTCTACTGTTGGCTGCAC
RM 11	7	(GA)17	TCTCCTCTTCCCCCGATC	ATAGCGGGCGAGGCTTAG
RM 30	6	(AG)9A (GA)12	GGTTAGGCATCGTCACGG	TCACCTCACCACACGACACG
RM 44	8	(GA)16	ACGGGCAATCCGAACAACC	TCGGGAAAACCTACCCTACC
RM 125	7	(GCT)8	ATCAGCAGCCATGGCAGCGACC	AGGGGATCATGTGCCGAAGGCC
RM 147	10	(TTCC)5 (GGT)5	TACGGCTTCGGCGGCTGATTCC	CCCCCGAATCCCATCGAAACCC
RM 163	5	(GGAGA)4 (GA)11C (GA)20	ATCCATGTGCGCCTTTATGAGGA	CGCTACCTCCTTCACTTACTAGT
RM 273	4	(GA)11	GAAGCCGTCGTGAAGTTACC	GTTTCCTACCTGATCGCGAC
RM 277	12	(GA)11	CGGTCAAATCATCACCTGAC	CAAGGCTTGCAAGGGAAG
RM 278	9	(GA)17	GTAGTGAGCCTAACAATAATC	TCAACTCAGCATCTCTGTCC
RM 283	1	(GA)18	GTCTACATGTACCCTTGTTGGG	CGGCATGAGAGTCTGTGATG
RM 287	11	(GA)21	TTCCCTGTTAAGAGAGAAATC	GTGTATTTGGTGAAAGCAAC

Genotyping protocol

Genomic DNA from each accession was extracted from fresh leaf tissues of 20-day-old plants according to a simple and modified protocol of Zheng *et al.,* 1995. The PCR was carried out in a reaction volume of 12.5 µl containing 5 to 25 ng of template DNA, 1.25 µl 10X PCR buffer without $MgCl_2$ (100 mM Tris-HCl pH 9.0 at 25°C, 500 mM KCl, 0.1% Triton® X-100 and H_2O), 1.5 µl of 25 mM $MgCl_2$, 0.25 µl of 10mM dNTP, 0.25 µl of 5 U/µl Taq polymerase enzyme, 0.625 µl each of 10 µM forward and reverse primers using a MJ Research single 96 well thermal cycler. To prevent evaporation, the mixture was overlaid with one drop of mineral oil. After an initial of 5 min denaturation at 94°C, each cycle consisted of 1 min denaturation at 94°C, 1 min of annealing at 55°C, and 2 min of extension at 72°C, with a final extension of 7 min at 72°C at the end of 35 cycles. The PCR products were then mixed with bromophenol blue gel loading dye and were analyzed by electrophoresis on 8% polyacrylamide gel using mini vertical polyacrylamide gels for high-throughput manual genotyping (CBS Scientific Co. Inc., CA, USA). 2.5 µl of amplification products were resolved by running gel in 1xTBE buffer for 2-2.5 hrs depending upon the allele size at a voltage of approximately 75 volts and 180 mA current. The gels were run with 0.5 mg/ml ethidium bromide stained and documented with UVPRO (Uvipro Platinum, EU) gel documentation device. Microsatellite markers or simple sequence repeat (SSRs) were used for DNA analysis (Temnykh *et al.,* 2001; McCouch *et al.,* 2002).

Data analysis

The size (in nucleotide base pairs) of the most amplified band for each microsatellite marker was determined by its migration relative to the molecular weight marker (50-bp DNA ladder) using Alpha Ease FC 5.0 software. SSR marker alleles were analyzed using Power Marker software. Summary statistics, including number of alleles per locus, major allele frequency, genetic diversity, polymorphism information content (PIC) values etc., were obtained using Power Marker software version 3.25 (Liu and Muse, 2005). Allele frequency data from the Power Marker software were used to export the data into a binary format, with presence scored as 1 and absence or missing observation scored as 0 of unique and shared bands for each marker allele-genotype combination for further analysis with NTSYS-pc version 2.2 (Rohlf, 2002). The similarity matrix was calculated with the Simqual subprogram using the Dice coefficient, then the cluster analysis was carried out with the SHAN subprogram using the UPGMA clustering method (UPGMA algorithm computed following 'Hierarchical cluster analysis') as implemented in NTSYS-pc version 2.2. For an unrooted phylogenetic tree, genetic distance was calculated using the 'Nei distance' (Rohlf, 2002), followed by phylogeny reconstruction based on the UPGMA method using neighbor-joining in Power Marker with tree viewed using TREEVIEW Win32 version 1.66 (Page, 1996). Finally, NTSYS-pc was used to construct a UPGMA (unweighted pair group method with arithmetic averages) dendrogram based on the Nei's (Nei and Takezaki, 1983) distance-based showing the genetic interrelationship among the genotypes. The genetic distance was calculated using the 'Nei genetic distance' (Nei, 1972; Rohlf, 2002).

RESULTS

Overall SSR diversity

Ninety-six rice landraces were successfully amplified with 12 SSR markers, where primer pairs called loci and DNA bands as alleles. A total of 252 alleles were detected in 12 SSR markers from 96 rice landraces. The size of the amplicons ranged from 42 to 165bp. The number of alleles per locus ranged from nine (RM163) to 41 (RM283) with a mean of 21. The largest range of band sizes was found for RM6 (102-165) followed by RM283 (80-160), RM273 (46-159), RM278 (87-158) and RM11 (57-137). Among the 30 SSR markers, the highest number of alleles (41) were found for RM283 followed by RM11 (25); RM6 and RM278 (24); RM44 and RM273 (21); RM125 and RM 277(18); RM30 and RM287 (17); RM147 (15); RM163 (11) (Table 3). The PIC values ranged from 0.759 (RM163) to 0.959 (RM283) with a mean of 0.759, indicating that the SSR primers used in this study were efficient and polymorphic. The PIC values for other markers were 0.93 (RM6, RM11), 0.92 (RM278), 0.91 (RM44), 0.90 (RM273), 0.89 (RM125, RM287), 0.87 (RM147, RM277), and 0.83 (RM30) respectively (Table 3). The PIC value revealed that RM283 was the best marker for 96 rice landraces. Figure 1 shows the DNA profiles of 96 Boro landraces with the SSR marker RM283.

Table 3. Allele number, allele size, frequency, genetic diversity and PIC for 12 SSR markers in 96 Boro rice landraces.

Marker	Chr . no	Position (cM)	Allele no.	Allele size (bp)	Allele frequency (%)	Genetic diversity	PIC value
RM 6	2	29.57	24	151.60	11.46	0.9397	0.9364
RM 11	7	47	25	120.88	10.42	0.9397	0.9364
RM 30	6	125.4	17	73	21.88	0.8520	0.8354
RM 44	8	2.88	21	97.09	13.54	0.9193	0.9137
RM 125	7	24.8	18	104	18.75	0.8932	0.8841
RM 147	10	20.68	15	87.53	22.92	0.8880	0.8788
RM 163	5	91.4	11	51.09	34.38	0.7867	0.7594
RM 273	4	94.4	21	119	14.58	0.9143	0.9080
RM 277	12	57.2	18	119.5	23.96	0.8850	0.8757
RM 278	9	77.5	24	140.79	13.54	0.9325	0.9286
RM 283	1	31.4	41	119.95	8.33	0.9609	0.9595
RM 287	11	68.6	17	106.47	17.71	0.8924	0.8830
Mean			21		17.62	0.9003	0.8916

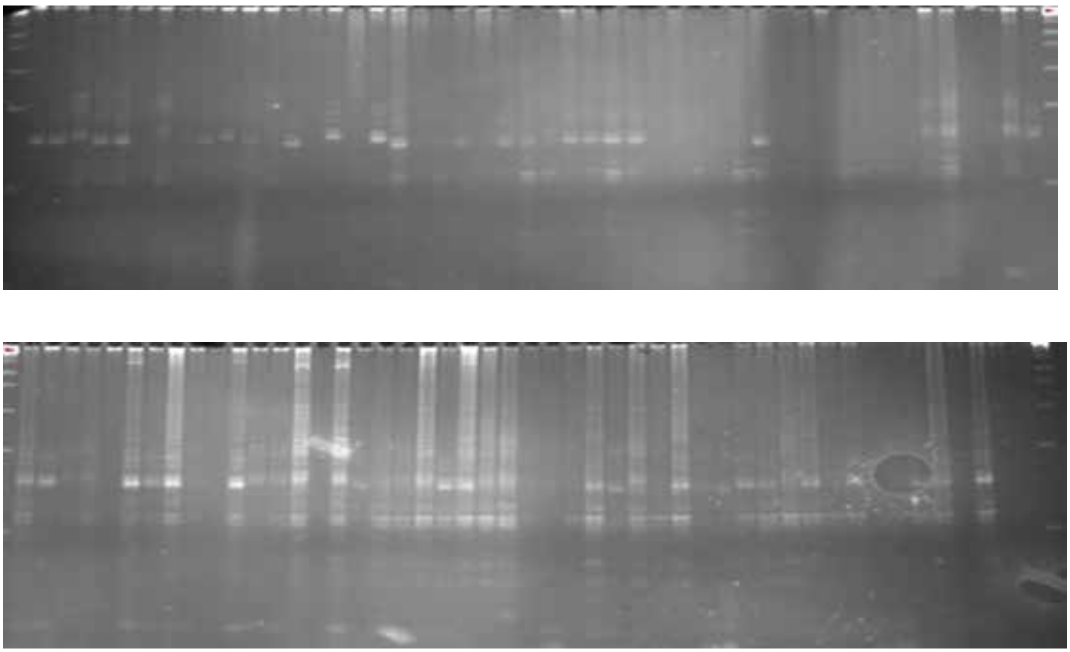

Fig. 1. DNA profile of 96 Boro landraces with RM283 (Number 1-96 represents rice landraces presented in Table 1).

Genetic distance analysis

Results based on genetic distance observed in the unrooted neighbour-joining tree produced seven clusters in 96 landraces (Fig. 2). The largest numbers of landraces (25) were found in cluster I, then in clusters VII (19), V (17), II and III (10), IV (9) and the least number in cluster VI (6).

UPGMA clustering was performed to group the landraces into a dendrogram. From this dendrogram, 96 rice landraces were classified into seven major clusters at a coefficient of 0.11, and the value of similarity coefficient ranged from 0.06 to 0.58. Cluster IV had 34 landraces and is the largest group of seven clusters, followed by cluster VI with 19 landraces; cluster II included 18 landraces; cluster I included 12 landraces; cluster III and VII included five landraces; cluster V included three landraces. The genetic differences between the 96 landraces were greatest for most genotypes (Fig. 3).

Genetic similarity analysis using the UPGMA cluster model was different for the landraces tested compared to that of unrooted neighbour-joining tree.

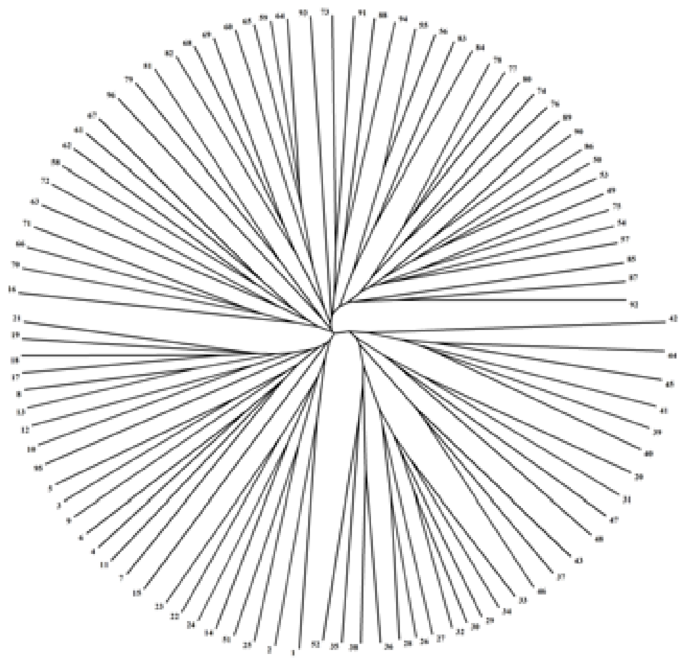

Fig. 2. An unrooted neighbour-joining tree showing the genetic relationships among 96 Boro rice landraces in Bangladesh (Number 1-96 represents rice landraces presented in Table 1).

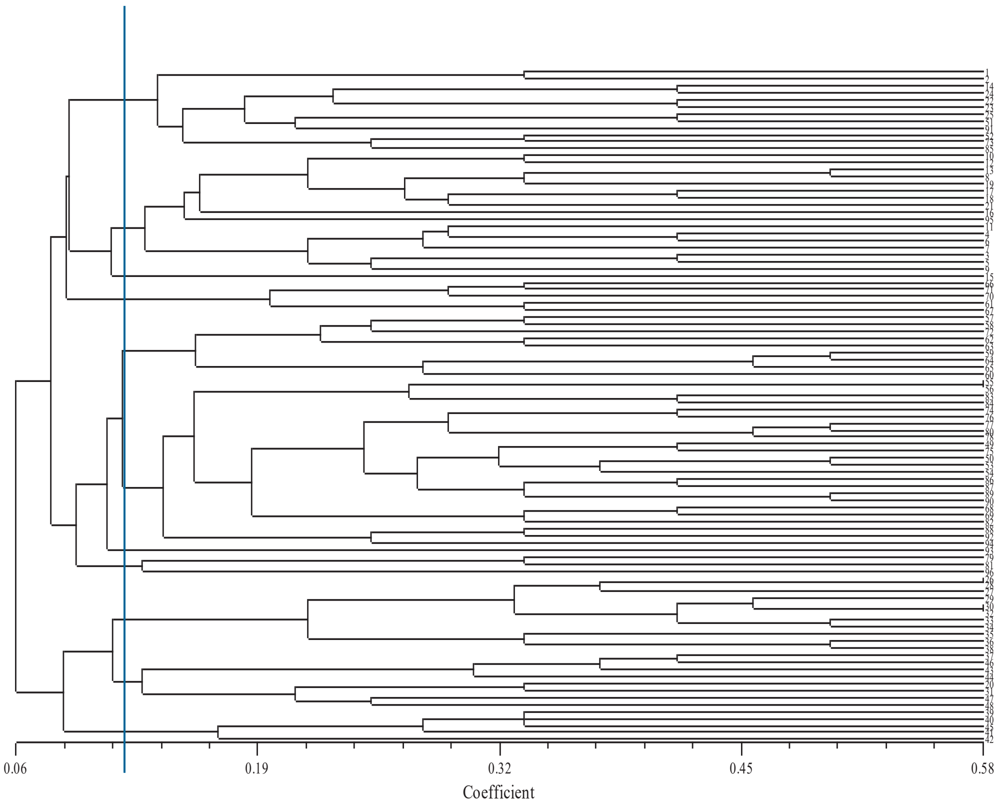

Fig. 3. Dendrogram of UPGMA cluster showing the genetic relationships between 96 Boro rice landraces of Bangladesh based on the alleles detected by 12 SSR markers (Number 1-96 represents rice landraces presented in Table 1).

Principal coordinate analysis

Two-dimensional and three-dimensional graphical views of principal coordinate analysis (PCoA) showed the spatial distribution of landraces along two and three principal axes. The landraces Bhaturi, Dholi boro, Boro, Ausha boro, Mukta har, Bati boro, Jamir, Kali boro, Khoea, Ulia, Solai, Isamoti, Lakhai, Panpiaj, KN-1B-361-1312-27-1, Ashani, Sona rata, Nata boro and Muirol were found far from the center of the cluster, while the remaining landraces were found approximately around the centroid. (Fig. 4 and 5).

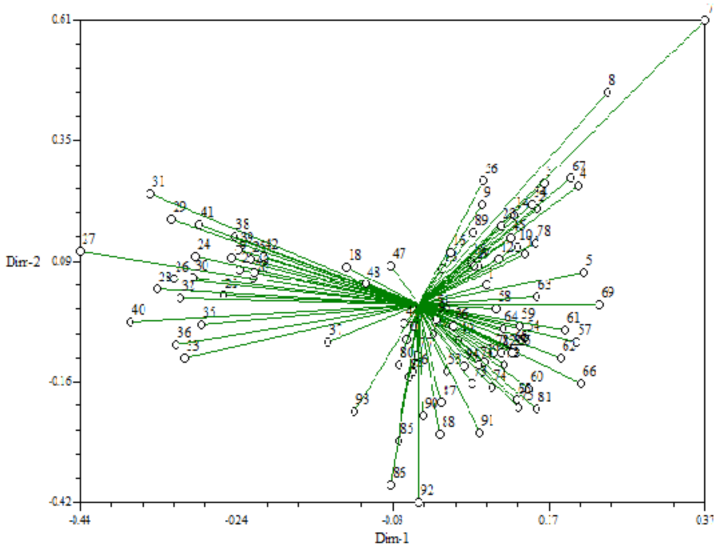

Fig. 4. Two-dimensional principal coordinate analysis (PCoA) view of 12 SSR markers in 96 Boro rice landraces (Number 1-96 represents rice landraces presented in Table 1).

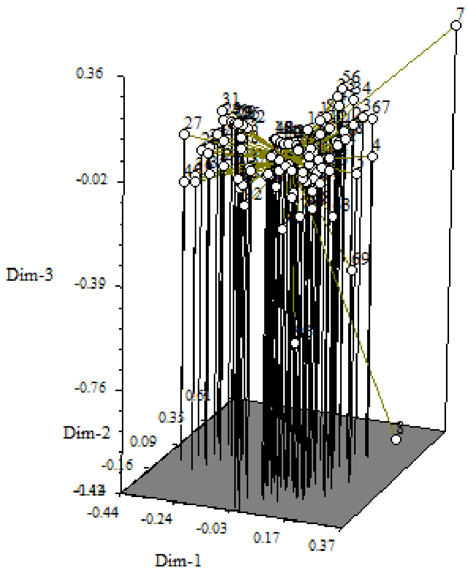

Fig. 5. Three-dimensional principal coordinate analysis (PCoA) view of 12 SSR markers in 96 Boro rice landraces (Number 1-96 represents rice landraces presented in Table 1).

DISCUSSION

The study of genetic diversity is of utmost importance in applied plant breeding to select parental genotypes in a plant breeding programme. Effective germplasm evaluation provides a scientific basis for parents/donor selection for varietal improvement, and selection of genotypes for specific agro-ecological conditions (Kumar *et al.,* 2012). Analysis of diversity at the molecular level using PCR-based markers is considered as an efficient and rapid method for identifying the associations and/or differences between genotypes (Schulman, 2007). Microsatellites are becoming increasingly popular and are suitable for large-scale analyses of genetic diversity and breeding studies (Brown and Kresovich, 1996; Joshi *et al.,* 2000). Competent and consistent use of molecular markers such as SSRs to study the genetic diversity of any food crop requires the selection and use of primers that provide clear, distinct, reliable and sufficient information required to study divergence within a crop (Arolu *et al.,* 2012). In our studies, the number of polymorphic loci detected by the primer combination varied depending on the primer. SSR markers amplified distinct band patterns in 96 Boro rice landraces, and each marker showed polymorphism.

The observed PIC values ranged from 0.759 (RM163) to 0.959 (RM283) and the mean was 0.759, which is consistent with the findings of Siddique *et al.,* (2014), who estimated genetic diversity in T. Aman rice germplasm collections using SSR markers, where PIC ranged from 0.65 to 0.91. The allele frequency ranged from 8.33% (RM283) to 34.38% (RM163) with an average of 17.62, which is consistent with the results of Siddique *et al.,* (2016d), who estimated the genetic diversity of the T. Aman rice germplasm collections using SSR markers, ranging from 8.33% to 22.92% with an average of 15.89. Gene diversity varied from 0.78 to 0.96 and its average value was 0.90, which also indicated the presence of adequate genetic diversity (Table 3). This average value was higher than that of Zai-quan *et al.,* (2012) and Siddique *et al.,* (2016b). Improved determination of relationships between 96 accessions was enabled by UPGMA cluster analysis using SSR markers. Seven major groups were found with a similarity coefficient of 0.11 in which rice accessions were mostly clustered together (Fig. 3), and the similarity coefficient ranged from 0.06 to 0.58. Siddique *et al.,* (2016d) also observed seven groups of T. Aman rice, whose similarity coefficient ranged from 0.06 to 0.75. It should be noted that SSRs formed seven groups, but in the subgroup of IV, the number of accessions was the highest (34) and the reason can be explained by the fact that SSR markers can target a larger number of repetitive sequences, in particular in the centromeric region that can significantly influence the classification pattern (Parsons *et al.,* 1997). These clustering patterns confirm the reliability of SSR markers for the genetic diversity analysis among Boro rice landraces. The principal coordinate analysis (PCoA) showed that landraces that were far from the centroid were more genetically diverse, while landraces that were close to the centroid had similar genetic backgrounds. Hence, the cluster analysis was largely supported by PCoA.

CONCLUSION

The results obtained in this study provided useful implications for establishing the sovereignty of the gene pool of Boro rice landraces in Bangladesh. This study showed a high level of genetic diversity in Boro rice, suggesting that SSR markers were very effective in detecting polymorphism in this ecosystem. To expand the genetic base and improve Boro rice, landrace with the lowest genetic similarity can be selected as parents. Hybridization must, therefore, be carried out between the genotypes of two distant populations. Taking into account all these criteria and the results of diversity analysis based on markers that are distant from each other due to their genetic relatedness (like Pashusail and Tulsi boro; Raja sail and Kali boro; Bashful and Jamir; Begun bitchi and Boro deshi; Banjira and Bogra (Deshi); Jagli boro and Lahi boro; Bimion and Gorchi sail; Jhati sail and Khaia boro; Tepi boro and Jamir boro) can be selected as parents for a further breeding programmes. This will lead to generate greater diversity and prebreeding lines will be developed with greater variabilities. Hopefully, this endeavour will increase genetic diversity in non-elite lines which will ultimately be helpful for achieving accelerated rate of genetic gain for different traits in breeding programmes.

AUTHORS' CONTRIBUTION

MAS and MK generated the idea; MAS and MZI developed methodology; MFRP, MAS, MHKB and AB gathered data; ESMHR and MSA supervised the study; MAS carried out analysis and wrote the manuscript; and PSB reviewed the manuscript.

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DECLARATION OF INTERESTS

The authors wish to confirm that there are no known conflicts of interest concerning this publication.

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Optimizing Hormonal Effects and Incubation Periods on In Vitro Regeneration in High-Yielding Indica Rice

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ABSTRACT

The research aimed to evaluate the effects of hormones and callus age on *in vitro* regeneration as well as seeking to establish a reliable and effective plant regeneration protocol for the rice varieties BRRI dhan92 and BRRI dhan96. Mature seeds were used to initiate callus induction using MS media containing 2 mg/L of 2,4-D. For the regeneration process, calli were transferred to MS medium supplemented with different hormone combinations: H1 (2 mg/L BAP + 1 mg/L NAA + 1 mg/L Kinetin), H2 (4 mg/L BAP + 0.5 mg/L NAA + 1.2 mg/L Kinetin), and H3 (1 mg/L BAP + 1 mg/L NAA + 1 mg/L Kinetin). Significant variations were observed in the response to plant hormones between the rice varieties. Additionally, calli of ten, fifteen, and twenty-one day old were tested to observe the effect of callus age on plant regeneration. The highest callus induction frequency was recorded in BRRI dhan92 (88.17%). The H2 hormone combination showed the highest regeneration frequencies, achieving 63.67% in BRRI dhan92 and 45.33% in BRRI dhan96. Furthermore, the highest regeneration frequencies were found in ten-day-old calli, with BRRI dhan92 at 67.00% and BRRI dhan96 at 49.00%. This optimized regeneration protocol for BRRI dhan92 and BRRI dhan96 can be effectively used for *Agrobacterium*-mediated genetic transformation and their subsequent improvement.

Key words: Incubation days, Hormone, *In vitro,* Regeneration, Callus induction.

INTRODUCTION

About one-third of the global population relies on rice (*Oryza sativa* L., 2n = 2x = 24), a member of the Graminae family and subfamily Oryzoidea, as their primary food source. Remarkably, rice also occupies about one-fifth of the world's cereal-cultivated land. Today, a staggering 90.5% of the world's total rice production comes from Asia, with significant contributions from China, Indonesia, Pakistan, India, Vietnam, Thailand, Myanmar, Bangladesh, the Philippines, and Japan (FAO, 2022). In Bangladesh, rice is the cornerstone of agriculture, accounting for half of the country's agricultural GDP and one-sixth of its total income. As the third-largest rice producer globally, Bangladesh dedicates approximately 11.70 million hectares to rice cultivation (FAO, 2022). With the population expected to soar to 215.4 million by 2050, the country will require 44.6 million tons of clean rice (Kabir *et al*., 2015). Although rice production in Bangladesh has quadrupled over the past five decades, the increasing population growth rate (1.22%) and diminishing arable land (-0.69%) (Khalequzzaman *et al*., 2005) necessitate further enhancements in rice productivity. To meet this growing demand, biotechnology, particularly genetic engineering and tissue culture, can be synergistically applied alongside traditional breeding methods

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(Binte Mostafiz and Wagiran, 2018). Genetic engineering and tissue culture present potent solutions to the limitations of conventional breeding, offering the potential to develop rice varieties with enhanced quality, yield, and resilience to both abiotic and biotic stresses (Tuteja *et al*., 2012).

Successful *in vitro* plant regeneration is paramount for leveraging these technologies to boost rice yields (Alam *et al*., 2012). An efficient *in vitro* regeneration protocol is crucial for the successful application of genetic engineering in developing transgenic rice varieties. However, plant regeneration through tissue culture is challenging across different rice varieties, with success heavily dependent on reliable callus culture and regeneration procedures. This technique has been pivotal in enhancing the quality and yield of desired plants (Ghorpade *et al.,* 2012).

A significant barrier to genetic modification of many plant species is the lack of an effective *in vitro* regeneration method (Azizi *et al*., 2015). Many agronomically desirable rice varieties exhibit resistance to *in vitro* regeneration due to inadequate callus development and regeneration capabilities (Khatun *et al*., 2003). Particularly, many indica rice cultivars are less susceptible to genetic alteration owing to their limited regeneration abilities. Factors influencing regeneration efficiency include genotype, type and physiological status of explants, culture medium composition, plant growth regulators, incubation period, and culture conditions, with genotype and nutrient composition being the most impactful (Mohiuddin *et al*., 2006).

Since 1994, BRRI dhan28 and BRRI dhan29 have been mega rice varieties for Boro season. Recently, they have been replaced by BRRI dhan96 and BRRI dhan92, respectively. These high-yielding BRRI varieties, were selected for this experiment due to their outstanding significance on yield performance. The objective was to identify the most effective hormonal combination and optimal incubation period for *in vitro* regeneration. Since there is no available tissue culture protocol of these newly developed rice varieties for transformation work, development of an efficient *in vitro* regeneration protocol is a top priority for the genetic transformation/genome editing and subsequent improvement.

Therefore, this experiment was conducted to evaluate the effects of various hormone combinations on the regeneration frequency of BRRI dhan92 and BRRI dhan96 to identify the most effective combinations. Subsequently, the selected hormone combination was used to assess the impact of callus age on regeneration capability. This research aims to develop a successful regeneration protocol for these rice varieties while minimizing experimental time in tissue culture.

MATERIALS AND METHODS

Seed collection Seeds of BRRI dhan92 and BRRI dhan96 were collected from the Biotechnology Division, Bangladesh Rice Research Institute in Gazipur.

Seed sterilization Eight hundred mature seeds of each variety were manually dehulled and subjected to a sterilization process in a laminar air flow cabinet. First, the dehulled seeds were sterilized with 70% ethanol for one minute and then rinsed with sterile water. Next, they were treated with 50% Clorox (v/v) containing one drop of Tween 20 for 20 minutes, with gentle agitation. To remove the sodium hypochlorite, the seeds were washed five to six times with sterile distilled water. Subsequently, the seeds were sterilized again with 50% Clorox (v/v) without Tween 20, followed by another five washes with sterile water to ensure complete removal of sodium hypochlorite. Finally, the sterilized seeds were placed on sterile filter papers to remove excess water.

Media Preparation, Observations, Collection and Scoring of data

The sterilized seeds were then placed into a callus induction medium (CIM) consisting of MS media (Murashige and Skoog, 1962) with 2.0 mg/L of 2, 4-dichlorophenoxyacetic acid (2, 4-D). Sucrose at 30 g/L used as the carbohydrate source, and phytagel at 4 g/L was the solidifying agent. The pH of the media was adjusted to 5.8, and it was autoclaved at 15 PSI and 121°C for 20 minutes. Six hundred sterilized seeds of each variety were placed on the callus induction media, and all culture plates were incubated at 25±1°C in the dark to induce callusing. Data on callus induction percentage were recorded 21 days after seed plating.

Three-week-old calli were then transferred into magenta boxes containing regeneration media supplemented with different hormone combinations: H1 (MS + 2 mg/L BAP + 1 mg/L NAA + 1 mg/L Kinetin), H2 (MS + 4 mg/L BAP + 0.5 mg/L NAA + 1.2 mg/L Kinetin), and H3 (MS + 1 mg/L BAP + 1 mg/L NAA + 1 mg/L Kinetin). These calli were maintained under a 16/8-hour light/dark conditions. Data on regeneration percentage and the number of green plants produced were collected.

Callus induction frequency (%) was calculated as follows:

$$\frac{\text{Number of embryogenic calli}}{\text{Total number of calli}} \text{X } 100$$

Regeneration frequency (%) was calculated as follows:

$$\frac{\text{Number of regenerated calli}}{\text{Number of calli incubated}} \text{X } 100$$

Another experiment was conducted to observe the effect of incubation period on regeneration frequency. The same seed sterilization and media preparation procedures mentioned above were followed. Calli aged ten, fifteen, and twenty-one days were transferred to regeneration media using the H2 hormone combination, as it showed the highest regeneration rates. The regenerated plantlets were successfully rooted on half-strength MS medium and later transferred into pots containing soil for acclimatization

RESULTS AND DISCUSSION

Callus induction The aim of the present study was to develop an effective *in vitro* regeneration protocol for the rice varieties BRRI dhan92 and BRRI dhan96. In this experiment, 2 mg/L of 2,4-D was used for callus induction. As a potent synthetic auxin, 2,4-D is often sufficient to initiate and sustain embryogenic callus growth in rice, hence its exclusive use at a concentration of 2 mg/L. Previous studies, including those by Shahsavari *et al*. (2010) and Hoque and Mansfield (2004), have shown that 2 mg/L is optimal for callus induction. This was further corroborated by Htwe *et al.* (2011). Figure 1 illustrated the callus induction frequency from dehisced seeds of both rice varieties. Callus induction in rice is highly variable and genotype-specific. Similar result was achieved by Joya *et al* (2019). Among the two varieties, BRRI dhan96 exhibited poorer callus induction compared to BRRI dhan92. The highest callus induction frequency was observed in BRRI dhan92 (88.17%), followed by BRRI dhan96 (71.61%). This difference highlights the genotype-dependent nature of callusing efficiency, as confirmed by Rashid *et al*. (2003), who reported significant variations in callusing among rice varieties. A prior study also indicated that tissue culture generates a wide range of variation correlated with incubation time and cultivar-specific factors (Rasheed *et al.,* 2005).

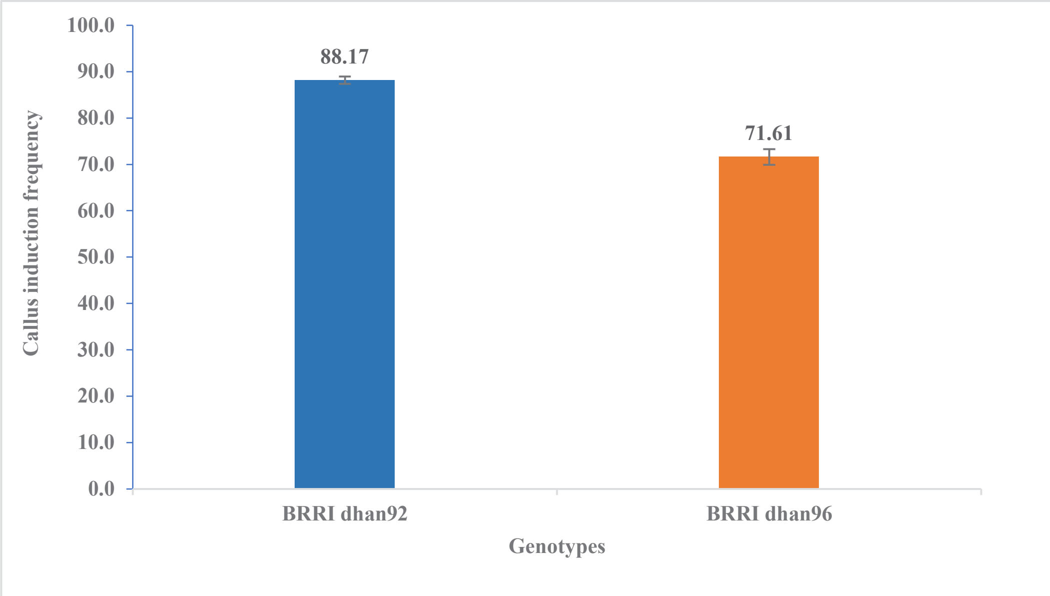

Fig. 1. Effect of genotypes to callus induction frequency.

Effect of hormone on regeneration frequency (%): Calli produced on MS media supplemented with 2.0 mg/L 2,4-D were evaluated for their plant regeneration ability under three regeneration media with different hormone combinations: H1 (2 mg/L BAP + 1 mg/L NAA + 1 mg/L Kinetin), H2 (4 mg/L BAP + 0.5 mg/L NAA + 1.2 mg/L Kinetin), and H3 (1 mg/L BAP + 1 mg/L NAA + 1 mg/L Kinetin). BRRI dhan92 exhibited the highest regeneration ability in H2 (63.67%) and the lowest in H3 (30.00%) (Fig. 2). Conversely, BRRI dhan96 also showed the highest regeneration ability in H2 (45.33%) and the lowest in H3 (23.67%) (Fig. 2). BRRI dhan92 generally responded better to all tested regeneration media compared to BRRI dhan96. This disparity may be attributed to the genotype and culture environment, as noted by Hoque and Mansfield (2004). Among the three media, the highest regeneration was observed in H2 and the lowest in H3. The study found that regeneration efficiency is influenced by varying concentrations of BAP, NAA, and Kinetin. The highest regeneration frequency was achieved with a high concentration of BAP (4 mg/L) and a low concentration of NAA (0.5 mg/L), similar to the findings of Tariq *et al*. (2008). A low concentration of Kinetin has been reported to enhance embryogenic calli and shoot formation efficiency in indica rice (Nhut *et al*., 2000; Humera and Jafar, 2011). Another study confirmed that successful plant regeneration is largely dependent on auxins and cytokinins combinations (Lee *et al*., 2002).

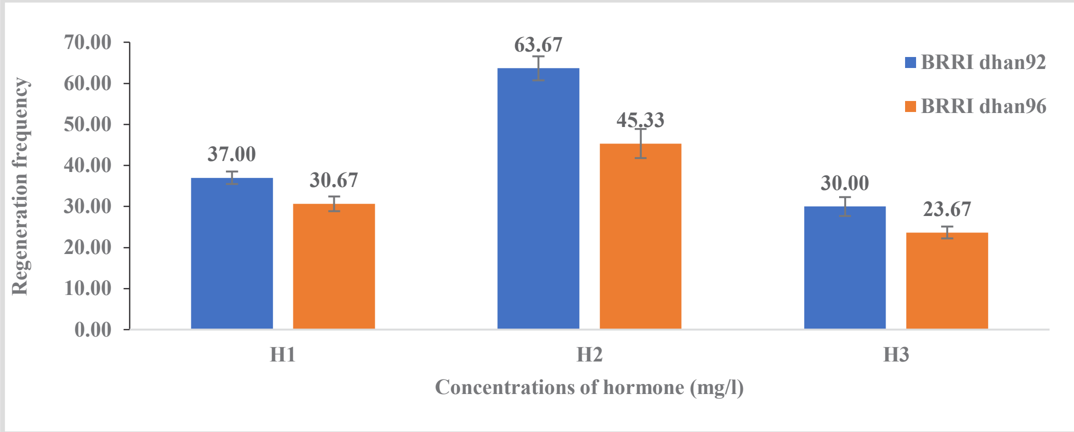

Fig. 2. Effect of different concentrations of hormone to regeneration frequency of BRRI dhan92 and BRRI dhan96.

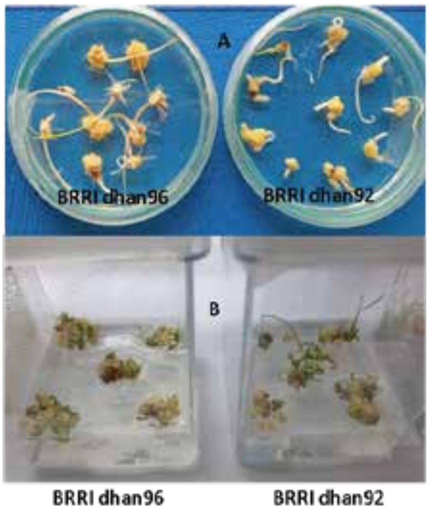

Fig. 3. Comparison of callus induction (A) and plant regeneration (B) between BRRI dhan92 and BRRI dhan96.

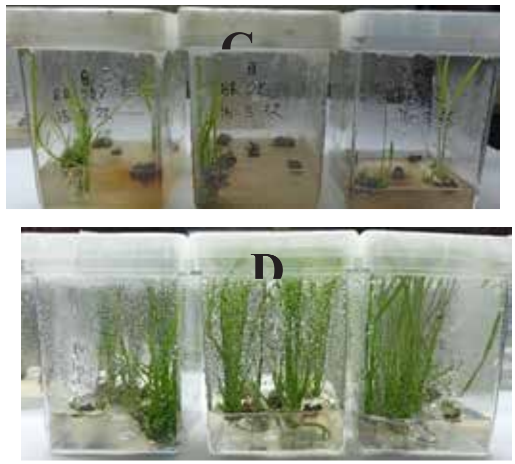

Fig. 4. Hormonal response to plant regeneration of BRRI dhan92; C= H1 and D= H2.

Effect of incubation days to callus induction frequency (%): BRRI dhan92 showed callus induction frequencies of 85.83%, 84.16%, and 83.33% at ten, fifteen, and 21 days of incubation, respectively (Table 1). In contrast, BRRI dhan96 exhibited callus induction frequencies of 72.33%, 68.50%, and 66.67% at the same respective intervals (Table 1). These results may be influenced by genotype and different incubation periods. Previous research has confirmed that tissue culture produces a wide range of variance due to the incubation period and cultivar specificity (Rasheed *et al.,* 2005). The experiment suggested that ten-day-old calli could be transferred to regeneration media, thereby reducing the time frame of the experiment.

Table 1. Effect of incubation days to callus induction frequency.

Days of transfer	Variety	Callus induction frequency (%)
Ten days	BRRI dhan92	85.83
	BRRI dhan96	72.33
Fifteen days	BRRI dhan92	84.16
	BRRI dhan96	68.50
Twenty-one days	BRRI dhan92	83.33
	BRRI dhan96	66.67

Effect of calli age on regeneration: In this experiment, calli aged ten, 15 and 21 days were transferred to the H2 hormone combination (H2 = 4 mg/L BAP + 0.5 mg/L NAA + 1.2 mg/L Kinetin) to observe the effect of callus age on plantlets regeneration. This experiment aimed to not only establish a regeneration protocol but also to minimize the experimental duration. The highest regeneration frequency (67.00%) for BRRI dhan92 was found in ten-day-old calli, with the lowest (62.00%) observed in 21 day-old calli (Table 2). The maximum number of regenerated plants (150) was also found in ten-day-old calli. For BRRI dhan96, the maximum regeneration frequency was 49.00% from ten-day-old calli, and the lowest was 44.00% from 21 day-old calli. The highest number of regenerated plants (60) was from ten-day-old calli, while the lowest number (47) was from 21 day-old calli (Table 2). A study highlighted that prolonged incubation of rice calli led to decreased regeneration efficiency due to increased oxidative stress and hormonal imbalances (Ali *et al.,* 2013). Another study also found that calli incubated for prolonged periods showed reduced regeneration efficiency due to decreased cellular viability and increased ROS levels (Mishra *et al*., 2020). From this phase of the experiment, it became clear that greater numbers of plantlets can be derived from ten-day-old calli. The insight of this study can significantly reduce the experimental time required in tissue culture.

Table 2. Effect of incubation days to plantlet regeneration and number of regenerated plants.

Days of transfer	Variety	% Plantlet regeneration	Number of regenerated plants
Ten days	BRRI dhan92	67	150
	BRRI dhan96	49	60
15 days	BRRI dhan92	65	140
	BRRI dhan96	46	52
21 days	BRRI dhan92	62	120
	BRRI dhan96	44	47

CONCLUSION

An efficient and reproducible regeneration system is crucial for controlling molecular mechanisms and achieving stable genetic transformation. Key factors for successful regeneration include genotypes, tissue source of explants, combination and concentration of growth regulators, and culture conditions. Considering this, our experiment evaluated the effect of different hormonal combinations on the regeneration rate of the newly developed rice varieties BRRI dhan92 and BRRI dhan96. The highest regeneration response was observed in H2 media, followed by H1 and H3. Based on these results, we conducted a further experiment to identify the shortest time required for optimal regeneration. Calli aged ten, 15, and 21 days were transferred to H2 media, which had demonstrated the highest regeneration frequency for both varieties. Remarkably, the ten-day-old calli showed the highest regeneration rate in the H2 hormone combination. This optimized regeneration protocol will significantly contribute to the enhancement of nutritional quality, resistance to biotic stresses (e.g., disease and pest resistance), tolerance to abiotic stresses (e.g., drought and salinity), and the improvement of specific traits in BRRI dhan92 and BRRI dhan96. Furthermore, gene editing technologies like CRISPR-Cas9 can be integrated with these regeneration protocols to precisely modify specific genes through *Agrobacterium*-mediated genetic transformation, enabling the production of genome-edited plants within a shorter timeframe. We hope our study will contribute to food security and agricultural sustainability.

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How Tobacco Makes Room in Rice Based Cropping Systems of Bangladesh

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Abstract

In spite of hazardous nature, tobacco has a significant position in the cropping system of Bangladesh. The existing status and determinants related to tobacco cultivation in Bangladesh remain inadequately explored. A thorough survey among farmers was conducted in 2021 to assess tobacco cultivation. The total tobacco cultivation area in Bangladesh was documented as 49 thousand hectares, accounting for approximately 0.57% of the overall net cropped area. Tobacco is distributed in 14 cropping patterns and cultivation is concentrated in 45 Upazilas of 15 districts. Farmers consider tobacco farming as a business and as a guaranteed cash crop at a pre-declared price rate. Poor farmers explore the opportunity of family labour employment in tobacco production and processing. Farmers are driven towards engaging in the business by a range of incentives offered by tobacco companies, while the fluctuating market prices of winter vegetables serve as an additional motivator. Many farmers perceive the risks associated with tobacco as being at a negligible level. Minimizing uncertainties of traditional agriculture may turn some farmers from tobacco to traditional crops.

Keywords: Agriculture, Cash crop, Food security, Soil fertility, Sustainability.

INTRODUCTION

Tobacco (*Nicotiana tabacum*), is the common name of several plants in the genus *Nicotiana* of the family Solanaceae that are used for cigarette, cigar, cheroot, bidi, hookah, and chewing purposes. It contains nicotine, an alkaloid organic compound that occurs throughout the tobacco plant and especially in the leaves and constitutes about 5% of the plant by weight. Worldwide, more than 70 species of tobacco are found, but the chief commercial crop is *N. tabacum* and in some countries, the species *N. rustica* is also cultivated (Sarkar and Haque, 2001).

Tobacco is considered a cash crop in Bangladesh and is commonly cultivated in the winter season/rabi season. Although it is a cash crop, the term 'crop' with this remains a question because it has some negative and controversial issues. Apart from the fact that tobacco is not indigenous to Bangladesh its introduction in the biodiverse agrarian systems in this country can also be criticised as introduction of an 'alien invasive species'. They disrupt an agro-ecological system by dominating over other cultivated and uncultivated crops essential for life and livelihood of a community (Ali *et al.*, 2015).

Despite various negative issues and controversies, tobacco has a significant impact on the world economy. The global

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tobacco market in 2010 was estimated at US$760 billion, excluding China(BAT, 2018). The global revenues from tobacco taxes in 2013-2014 was approximately $269 billion.In China, cigarette manufacturing is one of the few profitable state-owned industries. In India, tobacco generates approximately 20 billion Indian Rupees (US$0.45 billion) of income per annum as a result of employment, income, and government revenue (FAO, 2019).

Statista (2018) estimates that in the U.S. alone, the tobacco industry has a market of US$121 billion,despite the fact the CDC reports that US smoking rates are declining steadily. In terms of health expenditures, cigarette smoking contributed to more than $225 billion (or 11.7%) of annual healthcare spending in the U.S. in 2014. Of the 1.22 billion smokers worldwide, 1 billion of them live in developing or transitional economies, and much of the disease burden and premature mortality attributable to tobacco use disproportionately affect the poor. While smoking prevalence has declined in many developed countries, it remains high in others, and is increasing among women and in developing countries. Between one-fifth and two-thirds of men in most populations smoke. Women's smoking rates vary more widely but rarely equal male rates (Amiri, 2021).

There are 132 tobacco-producing countries in the world. Among them, 20 countries are cited with their production, area coverage and yield in Table 1 (FAO, 2019).Worldwide 6096 thousand tonnes of tobacco are produced per year. China is the largest tobacco producer in the world with 2242 thousand tonnes of production volume per year. Brazil comes second with 762 thousand tones of yearly production. India stands for the 3rd position in production though its area coverage is bigger than that of Brazil. In respect of tobacco production, Bangladesh stood in the 14th position whereas it has occupied 15th rank in area coverage of tobacco.

Although the government of Bangladesh earns significant revenue from Tobacco, the increasing trends in its cultivation are a great threat to the country's food security and sustainable crop production systems. It has been introduced in this country in the mid-sixties of the last century into the fields where food crops were grown, and more widely after liberation in 1971 by the British American Tobacco Company (Sarkar and Haque, 2001). Although Bangladesh Agricultural Research Institute (BARI) has conducted research and development activities on tobacco and was abandoned in 1995, tobacco production has mainly been pushed by big multinational companies such as British American Tobacco Company through contract growers (Sarkar and Haque, 2001).

Agriculture in Bangladesh is dominated by rice and sustainable intensification of rice-based cropping systems is very important for the country's food security. The average cropping intensity of the country is 200% of which 136% is contributed by rice (Nasim *et al.*, 2017). Rice is grown everywhere and throughout the year. The Winter season is the only time when diversified crops are accommodated in the rice-based cropping systems. In this winter season tobacco also fits itself in different cropping patterns. Tobacco is a non-food and hazardous crop. Despite many objections to its cultivation, the area of its cultivation is increasing in several locations of the country. Still there is no systematic study was conducted on this aspect in Bangladesh, and enough information is not available. Therefore, the study was carried out to investigate the current status of tobacco cultivation in Bangladesh, how it becomes a great threat to food security, and the reason why the area of its cultivation is increasing.

Table 1. World tobacco situation at a glance, 2018-19

Country	Area ('000 ha)	Prodn ('000 ton)	% of world prodn	Yield (ton/ha)	Prodn/ Person (Kg)
China	1004	2242	36.78	2.23	1.61
Brazil	356	762	12.5	2.14	3.64
India	418	750	12.3	1.8	0.56
United States	118	242	3.97	2.05	0.74
Indonesia	203	181	2.97	0.89	0.68
Zimbabwe	101	132	2.17	1.31	8.9
Zambia	66	116	1.9	1.77	6.87
Tanzania	163	107	1.76	0.66	1.97
Pakistan	46	107	1.75	2.3	0.53
Argentina	55	104	1.71	1.9	2.34
Malawi	86	95	1.56	1.11	5.32
Mozambique	79	94	1.54	1.18	3.25
North Korea	60	91	1.5	1.51	3.57
Bangladesh	42	89	1.46	2.1	0.54
Turkey	93	80	1.32	0.86	0.99
Thailand	20	67	1.1	3.43	0.97
Italy	17	59	0.97	3.45	0.98
Laos	6	54	0.89	9.52	7.76
Macedonia	17	26	0.42	1.54	12.31
United Arab Emirates	0.021	0.285	0	13.82	0.03
Other 112 countries	399	413	11.43	1.04	-
The world (132 countries)	3370	6096	100	1.81	0.79

Source: FAO, 2019.

METHODOLOGY

The study was conducted following two steps.

In the first step, a huge task was undertaken from August 2015 to November 2016 to identify the cropping patterns and their area coverage throughout the country. The study was carried out using secondary source of information from the Department of Agricultural Extension (DAE). DAE keeps records of crops in every nook and corner of the country. At the grass root level, in the blocks, Sub Assistant Agriculture Officer (SAAO) collects data from the field. They use the Mauza map to identify the net cropped area (NCA) and the land use for non-agricultural purposes. In the cropped area, major crop growing field and their coverage are also identified. SAAO surveys the possible ways to collect data. They collect the list of farmers and their lands from the manager of each Boro irrigation scheme. They also make a list of farmers for the other major crops like T. Aman, Aus, wheat, jute, etc. and minor crops as well. They collected data by interviewing farmers who cultivate different crops, and use different varieties in respective hectarages. He also uses his judgment by his eye estimation. Finally observing the actual harvest area, judging by his own experience, the individual crop, their varieties and coverage are identified. In these ways, they keep data of every crop and its coverage in their blocks. Combining block data, union, then upazila, then district data are compiled and kept in the record book of DAE. Thus in each Upazila, season-wise cultivated crops and their coverage are recorded.

DAE also keeps information on cropping patterns (CP). Each crop is cultivated in a specific field in a single, double, triple or quadruple CP. Based on individual crop coverage, the area of a crop in a CP is distributed. Other crops of these CP have to match in their respective coverage. Thus individual CP coverage data were developed.

Data we used in this study were collected from DAE. A semi-structured questionnaire was developed for the data collection on thecrop, CP, their area coverage, etc. for the year 2014. A small team of investigators visited the Deputy Director (DD), DAE office. They collected the secondary information of crops, and their hectarage from each Upazila from the district office. The questionnaires were then distributed to each Upazila to collect information on CPs and their hectarage and other related issues. Upazila Agriculture Office filled up the questionnaires and sent back to the investigators. These data were analyzed to find out the mismatch of data, if any, among the data and any queries regarding them.

Two sets of data, one collected from DD Office on crop and their coverage in each Upazilaand the other collected through the questionnaire on CP and their coverage from the Upazila Agriculture Office were analyzed. After analysis of the data, usually, there were some mismatches of data and some information that needed further clarification. To purify and finalize this information stakeholder consultation workshops were conducted in 64 districts separately to work on Upazilalevel data. A team of investigators (researchers) visited each district and organized a workshop. In the workshop, DD and district level all concerned officers of DAE and from Upazilalevel UAO, AEO, UAA, SAPPO, SAAOs of respective Upazila attended. In the workshop, if there is a mismatch of data or any other query on data, researchers pointed out it for discussion. Then the data were finalized.Thus the data used for the final analysis were the overviewed data. The CP for the present study with its hectarage means the proportion of areas under different CPs in each Upazila in 2014-15.

Collected data were analyzed using the Micro Soft Excel program. Tally, addition, average and descriptive statistics were used for the presentation of data. Finally, a detailed compilation of cropping patterns of the whole of Bangladesh was completed. From these massive data banks only the tobacco-based cropping patterns are extracted for this article.

In the second step, a socio-economic survey was conducted in 2021 to identify the causes and determinants for the continuous cultivation of tobacco. Five districts that are considered hotspots of tobacco production were selected for the study. The locations of the study were Mirpur and Daulatpur Upazilas of Kushtia district, Gangni Upazila of Meherpur district, Kaunia and Gangachara Upazilas of Rangpur district, Saturia and Sadar Upazilas of Manikganj district, and Lama Upazila of Bandarban district. From each district, 50 farmers were randomly selected. In the samples, all categories viz. company card-holder, non-card-holder, large farmers, medium, small and marginal farmers were included. Landless tobacco growers who cultivate tobacco on leased land were also included in the sample. Therefore, the total number of tobacco growers under interview was 250.

In order to collect desired information, an interview schedule was prepared to keep the objectives of the research in view. Farmers' opinion-based questions have been included in the schedule along with the selected characteristics of the respondents. It may be recalled that the schedules were pre-tested in an actual field situation before using the same for the final collection of data among 15 respondents (3 from each district) of the study area. Necessary corrections, additions and alterations were made in the interview schedule based on the results of the pre-test. Data were collected personally by the researcher themselves from the sample by using an interview schedule. Data collection was started on 22 May and completed on 30 June 2021 through semi-structured questionnaires. Data obtained from the respondents were transferred to the master sheet and then compiled to facilitate tabulation. The qualitative data were converted into quantitative ones by means of suitable scoring techniques. The analysis was done using SPSS (Statistical Package for Social Science) computer package. Descriptive analyses such as range, frequency count, number and percentage, mean, mode, median, and rank order were used.

RESULTS AND DISCUSSION

Position and distribution of tobacco in cropping systems of Bangladesh

In Bangladesh, 316 major cropping patterns were identified and among them, tobacco was found in14 cropping patterns (Nasim *et al.*, 2017). These cropping patterns were cultivated in about 49 thousand hectares of land which is equivalent to 0.572% of the net cropped area in the country (Table 2). The most dominant CP with tobacco was Tobacco-Jute-T.Aman which alone was covering 31% of the total tobacco area. The second and third dominant cropping patterns were Tobacco-Maize-T.Aman and Tobacco-Aus-T.Aman which had been occupying 15% and 12%, respectively of the country's tobacco area (Table 2).

Table 2. Distribution of tobacco in different cropping systems in Bangladesh, 2014-15

Cropping pattern	Area (ha)	% of total tobacco area	% of NCA in Bangladesh
01. Tobacco-Jute-T.Aman	15200	31.02	0.177
02.Tobacco-Maize-T.Aman	7470	15.24	0.087
03. Tobacco-Aus-T.Aman	6040	12.33	0.071
04. Tobacco-Fallow-T.Aman	5310	10.84	0.062
05. Tobacco-Jute-Fallow	4050	8.26	0.047
06. Tobacco-Aus-Fallow	3180	6.49	0.037
07. Tobacco-Boro-T.Aman	2300	4.69	0.027
08. Tobacco-Sesbania-T.Aman	1620	3.31	0.019
09. Tobacco-Vegetab-Vegetables	1500	3.06	0.018
10. Tobacco-Fallow-Fallow	1045	2.13	0.012
11. Tobacco-Maize-Vegetables	600	1.22	0.007
12. Tobacco-Sesbania	600	1.22	0.007
13. Tobacco-Mungbean-T.Aman	50	0.10	0.001
14. Tobacco-Mungbean-Vegetables	40	0.08	0.000
Total tobacco area	49005	100.00	0.572

Location-wise distribution of tobacco

Tobacco cultivation is distributed over 45 Upazilas in 15 districts (Figure 1). Therefore, it is clear that about one-fourth number of the districts are affected by tobacco cultivation. More specifically we can see that tobacco production is concentrated in about one-tenth number of Upazilas in the country. Among the 15 districts,Kushtia alone covers 17650 hectares of tobacco which is equivalent to 36% of the total tobacco area. Lalmonirhat district stands for the second position where 11990 hectares of land are covered by tobacco which occupies 24% of the total tobacco cultivation area of the country. These two districts together represent over 60% of the country's tobacco growing land. In consideration of individual Upazila it is distinct that the highest area (9200 ha) coverage of tobacco belongs to Mirpur Upazila of Kushtia district. This single crop accounts for the 39% land of net cropped area in the Upazila and this area is equivalent to about 19% of the total tobacco area.The follower Upazilas are Daulatpur of the same district, Aditmari and PatgarmUpazilas of Lalmonirhat district. They had 7720 ha, 4340 ha and 3120 ha of tobacco which represents about 16%, 9% and 6% share of the total tobacco area in the country, respectively. Therefore, one-half of the country's total tobacco area is concentrated in only these four Upazilas (Table 3).

Table 3. Location-wise distribution of tobacco cultivation in Bangladesh, 2014-15

Upazila	Tobacco area (ha)	% of respective NCA	% of total tobacco area in Bangladesh
01 Mirpur, Kushtia	9200	39.00	18.77
02. Daulatpur, Kushtia	7720	22.55	15.75
03 Aditmari, Lalmonirhat	4340	26.91	8.86
04 Patgram, Lalmonirhat	3120	14.92	6.37
05 Gangni, Meherpur	2700	11.34	5.51
06 Gangachara, Rangpur	2300	11.04	4.69
07 Kaliganj, Lalmonirhat	1820	9.35	3.71
08 Jaldhaka, Nilphamari	1750	7.39	3.57
09 Nilphamari Sadar	1700	5.62	3.47
10 Hatibandha, Lalmonirhat	1490	6.47	3.04
11 Lama, Bandarban	1300	13.66	2.65
12 Damurhuda, Chuadanga	1290	5.61	2.63
13 Lalmonirhat Sadar	1220	6.32	2.49
14 Alikadam, Bandarban	860	17.62	1.75
15 Manikganj Sadar	850	4.94	1.73
16 Naikhangchhari, Bandarban	700	8.74	1.43
17 Bheramara, Kushtia	610	6.74	1.24
18 Chakaria, Coxs bazar	600	2.78	1.22
19 Dighinala, Khagrachhari	600	7.48	1.22
20 Taraganj, Rangpur	600	5.21	1.22
21 Jhenaidah Sadar	500	1.86	1.02
22 Alamdanga, Chuadanga	400	1.34	0.82
23 Ramu, Coxs bazar	400	3.81	0.82
24 Shailkupa, Jhenaidah	350	1.16	0.71
25 Meherpur Sadar	300	1.40	0.61

Upazila	Tobacco area (ha)	% of respective NCA	% of total tobacco area in Bangladesh
26 Kishoreganj, Nilphamari	300	1.98	0.61
27 Baghaichhari, Rangamati	300	3.49	0.61
28 Domar, Nilphamari	250	1.25	0.51
29 Harinakund, Jhenaidah	200	1.36	0.41
30 Thanchi, Bandarban	170	4.55	0.35
31 Sundarganj, Gaibandha	150	0.48	0.31
32 Kushtia Sadar	120	0.52	0.24
33 Ruangchhari, Bandarban	110	3.96	0.22
34 Moheshpur, Jhenaidah	110	0.33	0.22
35 Daulatpur, Manikganj	110	0.77	0.22
36 Bandarban Sadar	80	1.65	0.16
37 Khagrachhari Sadar	80	1.52	0.16
38 Ghior, Manikganj	80	0.77	0.16
39 Matiranga, Khagrachhari	60	0.89	0.12
40 Mithapukur, Rangpur	50	0.11	0.10
41 Chuadnga Sadar	30	0.15	0.06
42 Kaliganj, Jhenaidah	30	0.14	0.06
43 Dimla, Nilphamari	30	0.13	0.06
44 Magura Sadar	20	0.07	0.04
45 Saltha, Faridpur	5	0.04	0.01
Bangladesh	49005	0.57	100.00

Socio-economic background of tobacco growers

The socio-economic characteristics and background of tobacco farmers influence their production to a great extent (Hassan*et al.*, 2001). In order to get a vivid picture of the socio-economic status of tobacco farmers, this paper includes age, education level, family size, main occupation status, types of farmers based on farm size, farming experience, tenurial status, and status of using communication devices.

Age

The socio-economic characteristics of the respondents surveyed on the selected tobacco farmer in the study area are presented in Figure 2. The results revealed that among 250 respondents, the age of the major (51%) tobacco farmers ranged from 36 to 55 years. About 27% of the respondent farmers were in the age group up to 35 years. Only 22% of farmers were in the age group above 55 years. It, therefore, indicated that a normal trend is prevalent here. Neither an attraction nor a negative attitude is extremely touching to a special age group.

Family size

It is observed from Figure 2 that, the majority (49%) of respondents have medium family size (4-6 members) followed by 32% of tobacco farmers having small family(1-3 members) and only 18% of the family belongs to a large family(more than 6 members). Since tobacco is a labor-intensive non-food crop, it is very helpful for tobacco farmers if the family size is larger.

Types of farmers

In the study area, as shown in Figure 2, the Majority of tobacco growers (53%) were from small land size groups (farm size 51-250 decimals). The second largest group

(24%) came from medium farmers. Landless (0-5 decimals) and marginal farmers (6-50 decimals) are also significant participants (17%) in tobacco cultivation. Only 5% of them are large farmers (farm size >500 decimals).

Educational status

Illiterate farmers or less educated farmers are generally more pursued in tobacco cultivation. Among the respondents, the maximum number of farmers (75%) are from the illiterate and primary education group. Eighteen percent of farmers had been having secondary education (Figure 2). Only 7% of farmers got a Higher secondary education level. It implies that more educated farmers have less tendency toward the production of tobacco in general.

Main occupation status

On the issue of main occupation status,only farming is the main source of earning for the majority (59%) of respondents in the study area.It is also observed that 30% and 8% of the tobacco growers earn mainly from business and service, respectively (Figure 2).

Tenurial status

The highest numbers of the respondents (42%) are those who grow tobacco in their own land. The second highest number of tobacco growers (40%) are hybrid model land owner (Fig. 1). They grow tobacco in their own land and in addition, they produce more tobacco in leased land as a profitable business item. The rest 18% are a tenant who grows tobacco in others' land received on the basis of a lease or mortgage system. Among the tenant group there are two subgroups. The first subgroup is composed of landless people. The second subgroup of people are the opportunist tobacco growers (6.8%). They have their own land, however, they do not grow tobacco in their land. They select more fertile and more suitable land from other people on the basis of lease or mortgage.

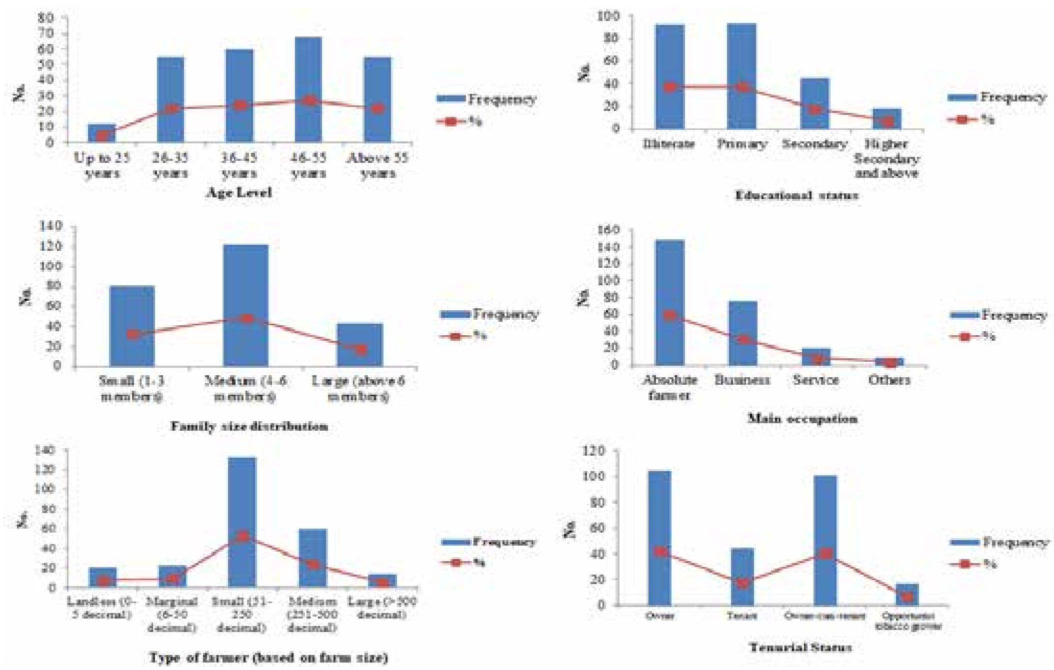

Fig. 1. Socio-economic characteristics of respondent tobacco farmers (N= 250)

Attachment of new farmers in tobacco cultivation

During the interview, a considerable point was recorded. That was about the experience of agricultural practices versus the duration of tobacco cultivation. Among the 250 farmers, 121 farmers had started their tobacco from their earliest time of agricultural farming. The rest 129 farmers started tobacco cultivation in the later stage. Descriptive statistics of these two series of 250 farmers have arranged for at a glance comparison (Table 4). Minimum value, average, mode and median all were bigger in agricultural farming experience than those of tobacco cultivation experience. This analysis detects that new farmers are coming to tobacco farming in the attraction of various advantages. In different studies, it was reported that tobacco extension workers from the multinational company go door to door for convincing the farmers with attractive proposals to join their tobacco stream (Motaleb and Irfanullah, 2011; Hossain and Rahman, 2013; Hassan*et al.*, 2001).

Table 4. Farmers' experiences in agriculture farming and tobacco cultivation

Experience (Years)	Minimum	Average	Mode	Median	Maximum
Agriculture farming	3	25.41	20	22.50	60
Tobacco cultivation	1	20.12	10	18.00	60
Difference (gap)	2	5.29	10	3.50	0

Ownership of tobacco farm and its cropping

In the previous section of this article, it was recorded that there is a good number of opportunist farmers. They are land-holders, however, they do not grow tobacco in their own land. They consider tobacco production as a profitable business so they grow it in others' land received based on lease or mortgage. The other group is landless people and they are obviously opportunists and they are bound to explore better land for better production of tobacco. They generally do not grow tobacco in consecutive years. They always try to select the land for the monoculture of tobacco. The second most important option for them is to keep the land fallow just before tobacco. For this reason, they try to select that land which is under flood water in kharif-II season. In the present study, the cropping status of own land and leased land is arranged to compare the cropping intensity (Fig. 2).

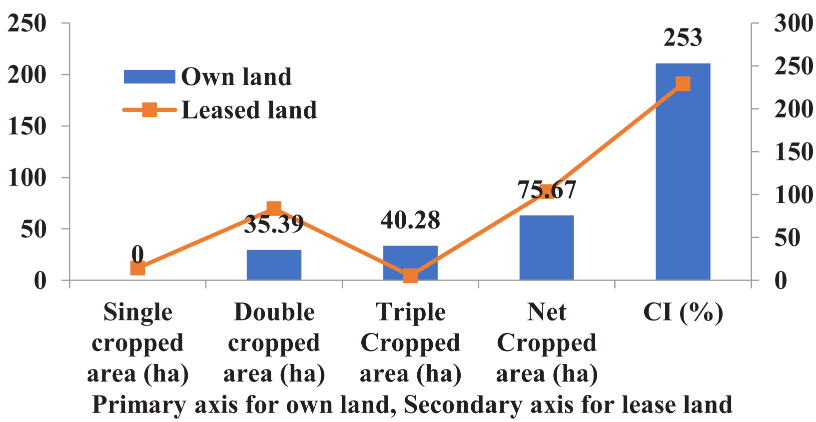

Fig. 2. Cropping intensity of tobacco growing farms in own land and leased land

In the list of own land, there was no single cropped area whereas a considerable area (11.94 ha) of leased land was under single cropped area. In the final calculation, cropping intensity was recorded 253% for own land and 191% for leased land (Fig. 3). Hence, it is proved that soil fertility is a crucial factor that is seriously considered by the farmers for tobacco cultivation. In several studies, there were some evidences and/or assumptions about soil fertility degradation (Akhter, 2018; Mollah, 2010).

Local variation of leasing value for tobacco cultivation

The highest average leasing value for tobacco cultivation is estimated as 84 thousand Bangladeshi taka per hectare followed by 77 thousand in Manikganj. This value was calculated at 75 thousand taka per hectare in Bandarban and 70 thousand in Meherpur. The least value was observed in rangpur and that was 25 thousand taka per hectare.

Ownership of tobacco farm and production economics

Production economics of tobacco is also highly affected by land ownership. The tenant farmers fix their target to get a maximum harvest of tobacco. So, their investment of capital and inputs was always higher than farmers who cultivate their own land. Use of urea, phosphate and potash fertilizer and total production cost of tenant farmers all were higher at a 1% level of significance than the respective values of own land farmers. Similarly, tobacco yield, gross return, and gross margin all were significantly higher at the 1% level than counter values under the cultivation of own land (Table 5).

Table 5. Input and output of tobacco cultivation in own land and leased land, 2021

Ownership category	Own land	Leased land	Difference
Use of fertilizer (kg/ha)			
Urea	244	315	71**
Phosphate fertilizer	213	297	84**
Potash fertilizer	111	167	56**
Yield (kg/ha)	2964	3567	603**
Production cost (‘000 tk/ha)	189.46	207.25	17.79**
Gross Return (‘000 tk/ha)	299.36	360.27	60.91**
Gross Margin (‘000 tk/ha)	109.90	153.02	43.12**

**Significant at 1% level by paired t-test.

Consciousness and ethical issues

The idea of ethics in tobacco farming differs from person to person. A tobacco farmer would consider not only personal cost-benefit but also social cost-benefit of it. He or she should consider the resulting short-run and long-run negative impact on public health, society and the environment. The nature of ethics would include awareness about tobacco causing individual health and public health hazards, the decline of soil fertility, environmental pollution, etc. and deciding on tobacco farming. To investigate whether farmers are known about the harms of tobacco farming and what is their comments about the validity of tobacco farming from an ethical ground, a summary of the opinion of the respondents about tobacco farming and its ethical related issues are described in Table 5. The majority of tobacco growers (57%) themselves are habituated to smoking and also in the use of *gul/jorda* (78%). It is shown in the findings that 43% of tobacco farmers in the study area say that it is harmful to health whereas 10% of them are not aware at all. Around 41% realize that tobacco cultivation pollutes the environment and 38% of respondents say that it leads to declining soil fertility gradually. Here it is implied that a significant number of farmers are not aware of tobacco-related environmental hazards and fertility decline. Successive queries about the negative impact, tobacco farmers are asked whether tobacco farming is unethical or not. In response of this question majority (57%) of them claim that tobacco farming is unethical (Table 6). A considerable number of farmers have no ethical concerns about tobacco farming. One-half of the respondents believe that tobacco is harmful for children.

Table 6. Opinion and perception of respondents about tobacco cultivation and some of the controversial issues, 2021

Opinions and awareness of the respondents	Yes	No	Not concern	Total
Smoker	141 (57%)	109 (44%)	-	250
User of Gul/Jarda	196 (78%)	54 (22%)	-	250
Tobacco production is harmful for health	107 (43%)	117 (47%)	26 (10%)	250
Tobacco cultivation pollutes environment	103 (41%)	122 (49%)	25 (10%)	250
Tobacco cultivation decreases soil fertility	94 (38%)	126 (50%)	20 (8%)	250
Tobacco cultivation is unethical	57 (23%)	99 (40%)	94 (38%)	250
Farmers should stop their tobacco cultivation	52(21%)	125 (50%)	73 (29%)	250
Harmful for children	125 (50%)	56 (22%)	69 (28%)	250

Factors pushing the farmers toward tobacco cultivation

The preference for farming tobacco is not merely an independent factor rather it is interlinked with many social, economic and individual factors. The study uncovers some of the causes that lead to the preference for tobacco farming. The most dominant cause of tobacco farming is its profitability over other crops. The other factors are mainly: having much money at a time, the opportunity of family labour employment, assurance of selling tobacco at a pre-declared fixed price, and so on (Fig. 4). A previous study also listed some determining factors which are responsible for the non-stop cultivation of tobacco. In the poor family, it is a business which is mainly based on family labors. Women and school going children are engaged in tobacco field operation, harvesting, curing and all other activities. Therefore, more family labor support leads a farmer to cultivate more tobacco. The findings are similar to the several studies(Bhavya, 2014; Abay*et al.*, 2004; Naher and Efroymson, 2007; Karagiannis and Sarris, 2005; Rahmanand Parvin, 2017; Hassan*et al.*, 2001; Kibwage*et al.*, 2009; Obwona, 2006; Ali*et al.*, 2015; Chikkala, 2015).

The factors shown in figure 4 are the determinants of the preference of tobacco cultivation. Among these factors, most of them are linked with the socio-economic consideration of the farmers. Here one factor is a very strong decider that is the short life cycle of the plant. In addition, it passes its seedling stage up to 50 days in the seedbed. As a result, its duration in the main field is again shorter than its total life span. In Bangladesh, cropping systems are extremely dominated by rice. In Kharif-II season (wet season) Aman rice is the only option. Tobacco can easily be transplanted after the harvest of Aman rice. If the Aman harvest is late, tobacco can also be transplanted in the rice field in a standing crop in a relay cropping system. These are the diversified ways through which tobacco is pushing itself in the cropping system without disturbing the preceding crop.

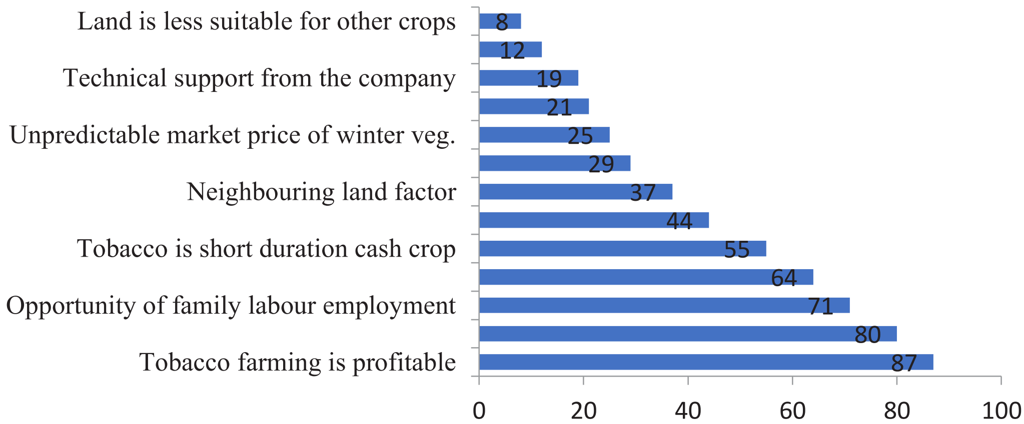

Fig. 4. Determinants of the farmers' preference for tobacco cultivation

CONCLUSIONS AND RECOMMENDATION

Farmers feel an interest towards tobacco due to short run profit. They can get a large amount of cash in one go. Apart from profit, many farmers are continuing tobacco with dissatisfaction especially for huge labour hours and health pains. Finding no other alternatives, many landless, marginal and poor farmers are entering into tobacco. Minimizing problems and uncertainties of traditional agriculture may turn some farmers from tobacco to traditional crops.

Until the market of traditional crops is ensured, there is less likelihood that farmers skip tobacco cultivation. In the study area, it was observed that there is no direct policy to regulate tobacco. As there are many long-run cost and negative externalities occurred for tobacco cultivation, there must be a policy balance between imposing control over tobacco companies and minimizing challenges of traditional agriculture.

Tobacco is not only a national concern, it is an international issue. It is a big trade in the world market. The governments of the countries are collecting huge revenue. National and international policy makers are also consuming tobacco without any hesitation. Therefore, it is a serious dilemma that might not touch the goal bar. However, the policy makers and researchers should make some avenues and alternatives so that unwilling poor farmers can get relief from unfair circle of tobacco. Some guidelines may be introduced as follows:

1. Conception of tobacco elimination should be discarded. Eradication of an item is a tough job and it may not be possible. Strong management policy should be adopted to keep it under a thresh-hold level.
2. Tobacco research should be empowered for ultra-high yielding varieties, so that vertical expansion will meet the demand and tobacco area will be reduced.
3. Scientific safety measures (as best as possible) should be ensured for tobacco growers and labourers.
4. Social awareness should be developed and some awareness

program should be arranged for the propagation of bad impact of tobacco.

5. The consciousness of the farmers about the negative impact of tobacco and its validity from ethical ground needs to be increased.
6. Necessary steps to be taken to ensure fair and stable price of food crops and minimizes the risk of damaging crops.
7. Initiatives to be taken to spread out the idea about ethical legitimacy of tobacco farming through educational institutions, religious institutions (i.e. Mosque, temple) and local Union council.

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Thermal Requirement for the Phenological Development and Yield of Rice as Influenced by Seeding Date

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ABSTRACT

The local climate greatly influences the development of a crop including factors such as temperature, rainfall, light intensity, radiation, and sunshine duration. Thermal units are closely linked to various physiological processes at each stage of crop development and can serve as a valuable tool for assessing thermal response. To explore this, a study was conducted to evaluate the grain yield and thermal unit requirements for the phenological development of four rice varieties under different seeding dates. Seeding took place from 15 November to 30 December, at 15-day intervals. The findings revealed that the seeding date had a significant impact on the thermal unit requirements for crop phenology and yield. Earlier seeding resulted in a higher accumulation of degree days compared to later seeding, as the crops required the most thermal input to complete panicle initiation. Seeding date notably affected the accumulated Growing Degree Days (GDD) from sowing to panicle initiation, heading, full flowering, and maturity. The GDD requirements for physiological maturity were the highest for the November 15 seeding, with the required GDD decreasing as seeding was delayed. The accumulated GDD for physiological maturity across the four varieties under different seeding dates ranged as follows: BRRI dhan96 (1972 to 2112 °C day), BRRI dhan97 (2116 to 2267 °C day), BRRI dhan99 (2097 to 2267 °C day), and BRRI dhan100 (2031 to 2183 °C day). Along with GDD, Heat Use Efficiency (HUE), Helio Thermal Units (HTU), and Photothermal Units (PTU) were identified as useful metrics for estimating crop growth and development based on temperature and solar radiation. In terms of yield, BRRI dhan96 achieved the highest yield when seeded on 30 November, outperforming the other three varieties (BRRI dhan97, BRRI dhan99, and BRRI dhan100). Therefore, the study concludes that seeding between 15 and 30 November optimizes heat use efficiency, leading to better physiological responses and higher yields.

Key words: Rice, Seeding date, Phenology, Growing degree days, Heat use efficiency

INTRODUCTION

Rice is the dominating and staple food crop in many Asian countries along with Bangladesh (Rao et al., 2007; Halder *et al*., 2016; Kumar and Ladha, 2011). It can grow in diverse environments from below sea levels to hill (Rami *et al.,* 2010). Different weather variable like rainfall, temperature and bright sun shine hours is the key to determine rice crop phenological development and yield. The date and time of specific development of a crop can be described by studying its phenology. It was reported that the growing degree day (GDD), helio-thermal unit (HTU) and photo-thermal unit (PTU) are some general parameters which can draw a relationship among plant growth and st environmental conditions like temperature, bright sunshine hours, and day length (Pandey *et al.*, 2020). The heat unit concept gives assessment of potential yield of a crop in different weather conditions (Kumar *et al.,* 2014). The growth and development of rice is controlled by temperature and photoperiod, however, the

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rate of growth for photo-insensitive rice is mostly depends on temperature. The amount of heat energy an organism accumulates over a period of time is often expressed as a `growing degree-day' (GDD). GDDs are used to relate plant growth, development, and maturity to air temperature and assesses the phenological events of rice of a particular region and crop growing season (Rajput et al. 1987). It also determines the crop growth stage and predict the best time for fertilizer and pesticide applications as well as estimates the heat stress and maturity and harvesting dates (Parthasarathi *et al.*, 2014). The efficiency of heat utilization in relation to crop yield referred to Heat use efficiency (HUE) which has a considerable application in crop field, depends on crop types and its genetic and environmental factors. Temperature directly influences the duration of crop growth stages, which could be predicted by using the aggregate of daily air temperature. So, it is crucially important to know the knowledge of exact span of various phenological stage of a crop grown in a particular environment and their consequences on yield (Dalton, 1967 and Wang, 1960).

Therefore, taking the above facts, the objectives of this study is to determine the optimum seeding date and yield with agro-climatic indices viz. GDD, HUE, HTU and PTU of four BRRI varieties.

MATERIALS AND METHODS

The field experiment was conducted at the research farm of Bangladesh Rice Research Institute, Gazipur in Boro 2021-22 season. It is situated at 23.99° N and 90.40° E. The experiment included four dates of seeding viz. 15 November, 30 November, 15 December and 30 December where transplanting was done at 40 days after seeding as main plot treatments and four varieties namely BRRI dhan96, BRRI dhan97, BRRI dhan99 and BRRI dhan100 as subplot treatments, resulting in 16 treatment combinations. The experiment was laid out in a split-plot design with three replications. Fertilizers were applied @74-60-70-35 kg ha^{-1} Urea-TSP-MoP-Gypsum respectively. All fertilizers were applied during final land preparation except N as a form of urea, which was top dressed in three equal splits at 15, 40 and 65 DAT (days after transplanting). The sub-plot size was 12 m^2 (3m × 4m) having a plot to plot and block to block distance of 0.75 m and 1.0 m respectively and there were 48 plots in the experiment. Weeds were controlled manually after 1st and 2nd top dress of urea. The different phenological stages like panicle initiation, first heading, complete flowering and physiological maturity of rice varieties were recorded by visiting the field from transplanting to harvesting. At maturity grain yield was measured, after discarding border areas on all four sides of the plot, harvesting 5m^2 area and after threshing and cleaning the grain weight was adjusted at 14% moisture by following method described by Gomez, 1972.

Data of weather parameters viz., maximum temperature, minimum temperature and bright sunshine hours during the experiment period were collected from agrometeorological observatory of Plant Physiology Division, BRRI, Gazipur and an automatic weather station (Davis Vantage Pro2 Weather Station) of Plant Physiology Division, BRRI. The Distance between agrometeorological observatory and experiment site is about 500 m. The weather data collected from automatic weather station were used to verify the GDD calculated from manual weather station and all the other indices data generated from manual weather station data were found with minimum variation.

Agrometeorological indices like growing degree days (GDD), helio thermal unit (HTU), photo thermal unit (PTU) and heat use efficiency (HUE) were calculated by using following formulas:

Growing degree days (by Nuttonson, 1948):

Growing Degree Days (GDD)=

$$\sum_{i-1}^{n} \left(\frac{(\text{T Max}+\text{T Min})}{2} - Tbase\right)$$

Where, T max and T min are the maximum and minimum temperature of the day and T base is the minimum threshold temperature of the crop, also called as base temperature or minimum threshold temperature. The base temperature of rice crop of 10 °C was used for computation of GDD on daily basis (Thomas, 1957).

The data on GGDs accumulation were further used to calculate heat use efficiency (HUE; Rajput, 1980) and helio thermal units (HTU; Satake, 1978) as under:

$$\text{Heat Use Efficiency (HUE)} = \frac{\text{Grain Yield } (\frac{\text{kg}}{\text{ha}})}{\text{GDD}}$$

$$\text{Helio Thermal Units} = \text{GDD} \times \text{Actual sunshines hours}$$

Photothermal units (PTU), degree day hrs

Photothermal units are the cumulative value of growing degree days, multiplied by the day length. This can be mathematically represented using the following formula (Dagar *et al.,* 2017):

$$\text{PTU} = \sum (\text{GDD} \times \text{N})$$

Where, GDD = Growing degree day, N = Maximum possible sunshine hours or day length (hrs)

RESULTS AND DISCUSSION

Four BRRI developed Boro rice varieties BRRI dhan96, BRRI dhan97, BRRI dhan99 and BRRI dhan100 were seeded at four different seeding windows starting from 15 November to 15 December at 15-days interval to find out the effect of different agro-climatic indices for different phenological development. During the experimental period the highest maximum temperature 37.5 ºC was recorded at April (Fig. 3) and lowest minimum temperature was recorded at February (10.6 ºC) whereas January was found the coolest month (average temperature was 20.11 ºC) and April was found hottest month (average temperature was 30.04 ºC). During March maximum sunshine hour and solar radiation were harvested by crop (daily average 7.57 hour and 16.95 MJ/day/sqm respectively).

Thermal time requirement for crop phenology

Table 1 presents the henology of four rice varieties under different seeding times. The results indicated that phenology was significantly influence by of seeding date. The numbers of days for attaining different phenological stages differed for seeding date. In case of 1st seeding (15 November), all the varieties require more number of days for attaining panicle initiation (PI), first heading and complete flowering compared to the other three seeding date. The higher growth duration occurred for 1st seeding because after transplanting compared to the others the plant faced lower temperature than critical low temperature for rooting and tillering (16ºC, Yoshida, 1981) for maximum periods (Fig 3). Which might delay the root growth and tiller production resulted the increased growth duration.

The panicle initiation (PI) of BRRI dhan97 required maximum (99) days, followed by BRRI dhan99 (96 days), BRRI dhan96 (89 days) and BRRI dhan100 (87 days). Thermal time required (GDD) from sowing to PI, heading, complete flowering and maturity was significantly affected by sowing date. The range of days to 1^{st} heading were found 96 to 119 days, 110 to129 days, 106 to 127 days and 97 to 117 days for BRRI dhan96, BRRI dhan97, BRRI dhan99 and BRRI dhan100 respectively. In general, 15 November sowing rice plants accumulated higher degree days and with each delay in sowing the degree day accumulation decreased during the crop seasons. Similar result was found by Dagar *et al.,* 2017 and Moti *et al.,* 2015. The highest GDD was accumulated from seeding to PI by the first (15 Nov.) seeding followed by second (30 November), third (15 December) and fourth seeding (30 Dec.) for all the variety except BRRI dhan97. Among the variety BRRI dhan99 required highest GDD to PI (1484 °C day) followed by BRRI dhan97 (1108 °C day), BRRI dhan96 (1005 °C day) and BRRI dhan100 (981 °C day) at 15 November sowing. Similarly, the highest thermal requirement was observed for heading and complete flowering for most of the varieties at 15 November sowing. Where, The GDD values for heading and complete flowering was the highest in BRRI dhan97 (1594 °C day and 1787 °C day respectively) followed by BRRI dhan99 (1555 °C day and 1767 °C day respectively) at 15 November seeding. BRRI dhan96 required highest GDD for first Heading (1407° C/day) but less GDD to complete flowering (1555 °C day) than BRRI dhan100, which GDD value for first heading and complete flowering was 1375 °C day and 1613 °C day respectively. The total GDD required for physiological maturity was maximum in 15 November seeding and it was gradually decrease with delay seeding up to fourth seeding for BRRI dhan96 and BRRI dhan100. Similar GDD value was found in BRRI dhan97 and BRRI dhan99 (2267 °C day) for physiological maturity and among the four seeding it was highest and the requirement of thermal unit decrease in delay seeding up to 15 December seeding then it was slightly increase at fourth seeding (30 December). The maximum heat unit of BRRI dhan96 and BRRI dhan100 was 2112 and 2183 °C day respectively at 15 November seeding. The reason behind the higher degree days of 15 Nov. and later on seeding time can be explained from table 3 where, we observed the higher GDD acquired by plant at seedling stage was higher in 1^{st} seeding then 2^{nd} seeding for most of the variety. The seedling of last two seeding faces cooler period than first two seeding. The range of GDD for seedling stage and transplanting to panicle initiation for all the variety was 345 to 389 and transplanting to panicle initiation stage it was 344 to 431, 480-556, 502 to 522 and 360 to 408 for BRRI dhan96, BRRI dhan97, BRRI dhan99 and BRRI dhan100 respectively. Here an interesting thing we observed that, the GDD calculated from automatic weather station data is quite lower than the manual weather station. This is because the manual weather station had only two data point but automatic weather station had 12 data point (every 30 minute) in each day and the most of the growing condition faced cooler period specially for the first two seedings. So the average maximum temperature was lower than the manual weather station. The cooler period delays the growth of first seeding causes higher growth duration and gradually enhance the growth as the temperature increased results of growth duration reduction with the advancement of season.

So, it was observed that, early seeded crop had higher degree days and with the delay in each seeding time the requirements of degree days decreased during the crop season mainly due to the GDD acquired by seedling stage. Similar result was reported by Sing and Paul, 2003 and Khan *et al.,* 2006. They reported that at wet season under advance transplanting the Growing degree days is higher and subsequently decreasing at delay transplanting.

They found that the GDD requirement for the maturity was the highest in Lal Swarna (2385 degree C/day), followed by IR-36 (1975 degree C/day) and Kshitish (1913 degree C/day).

The range of accumulated GDD reached physiological maturity under different seeding date from 15 November to 30 December of BRRI dhan96, BRRI dhan97, BRRI dhan99 and BRRI dhan100 was 1972 to 2112 °C day, 2116 to 2267 °C day, 2097 to 2267 °C day and 2031 to 2183 °C day respectively. Dagar *et al.,* 2017 observed for different transplanting dates, accumulated GDD ranged from 1791.0 to 2342.9 degree days, 1802.2 to 2133.9 degree days, 1802.2 to 2118.7 degree days and 1575.3 to 1903.6 degree days for varieties CSR 30, HB 2, PB 1121 and PB 1509 respectively.

Table 1. Days required for development of crop phenology of four varieties under different seeding date.

		Phenological stages			
	Seeding date	Day to panicle initiation	Day to first heading	Day to complete flowering	Growth duration (day)
BRRI dhan96	15 Nov	89	119	127	154
	30 Nov	80	110	124	142
	15 Dec	72	102	116	136
	30 Dec	66	96	108	129
BRRI dhan97	15 Nov	99	129	139	163
	30 Nov	86	116	130	151
	15 Dec	83	113	129	146
	30 Dec	80	110	123	143
BRRI dhan99	15 Nov	97	127	138	163
	30 Nov	89	119	132	152
	15 Dec	82	112	125	145
	30 Dec	76	106	116	142
BRRI dhan100	15 Nov	87	117	130	159
	30 Nov	81	111	126	149
	15 Dec	73	103	118	143
	30 Dec	67	97	108	135
CV%		2.28	1.67	1.37	5.82
LSD (0.05) for seeding date		1.55	1.15	1.42	1.71
LSD (0.05) for variety		1.55	1.55	1.42	1.71
LSD (0.05) for variety*seeding date		3.10	3.10	2.84	3.42

Table 2. Required total growing degree days (GDD) at different phenophases of four varieties under different seeding dates.

Variety		Growing degree day (°C)			
		15th Nov	30th Nov	15th Dec	30th Dec
BRRI dhan96	Panicle initiation	1005	852	742	713
	First heading	1407	1309	1249	1264
	Complete flowering	1555	1577	1524	1528
	Maturity	2112	1936	1926	1917
BRRI dhan97	Panicle initiation	1108	921	903	955
	First heading	1594	1422	1464	1548
	Complete flowering	1787	1678	1779	1806
	Maturity	2267	2120	2116	2188
BRRI dhan99	Panicle initiation	1484	965	888	882
	First heading	1555	1480	1444	1666
	Complete flowering	1768	1738	1708	1682
	Maturity	2267	2139	2097	2172
BRRI dhan100	Panicle initiation	981	864	757	728
	First heading	1375	1372	1269	1284
	Complete flowering	1613	1597	1564	1528
	Maturity	2183	2077	2056	2031

Table 3. Required total growing degree days (GDD) for different vegetative periods of four varieties under different seeding dates. (Data were taken from Davis Vantage Pro2 Automatic Weather Station) of Plant Physiology Division, BRRI).

Variety		Growing degree days (°C)			
		15th Nov	30th Nov	15th Dec	30th Dec
BRRI dhan96	Panicle initiation	919	784	705	690
	seedling	489	392	334	345
	TP to PI	431	392	371	344
BRRI dhan97	Panicle initiation	1039	872	866	902
	seedling	489	392	334	345
	TP to PI	550	480	532	556
BRRI dhan99	Panicle initiation	1009	914	851	847
	seedling	489	392	334	345
	TP to PI	521	522	517	502
BRRI dhan100	Panicle initiation	896	797	718	705
	seedling	489	392	334	345
	TP to PI	408	405	384	360

Helio Thermal Unit (HTU)

The variation in mean daily temperature and bright sunshine hour among four varieties cropping period under different seeding date resulted in varied accumulated helio-thermal units for life cycle of all of the varieties. The total helio-thermal units were observed for different seeding date ranged from 12,229 to 14,626 °C day hour. The data presented in show that total HTU required during total crop growth period was the highest in 15 November seeding for all of the varieties. BRRI dhan96 required maximum 13,607 °C day hour HTU at 15 November seeding followed by 30 December (12,645 °C day hour), 15 December (12,618 °C day hour) and 30th November seeding (12,229 °C day hour). BRRI dhan97 and BRRI dhan99 had similar HTU (14,226 °C day hour) at 15 November seeding and 15 December Seeding (13,792 °C day hour) as well the required HTU of BRRI dhan100 was maximum 13,902 °C day hour at 15 November seeding followed by 15 December (13,475 °C day hour), 30 November (13,307 °C day hour) and 30th December (12,870 °C day hour) seeding. In a study by Sultana *et al.,* 2019 the maximum helio-thermal units (15141.01 °C day hour) were recorded in BRRI dhan49 followed by (13759.75 °C day hour) in the advanced line BR (Bio) 9786-BC2- 119-1-1 and the lowest (10520.26 °C day hour) in the advanced line BR (Bio) 9786-BC2-119-1-3 at the 5th July Seeding. Mote *et al.,* (2015) observed the range of HTU of rice variety cv. Jaya were 11784 to 12990.7ºC day hr.

Heat Use Efficiency (HUE)

Heat use efficiency (HUE), is the conversion of heat energy into dry matter production and depends on crop type, genetic factors and sowing time (Rao *et al.,* 1999). Total heat energy available to any crop is never completely converted to dry matter even under most favorable agro climatic conditions. Among the seeding dates 30 November seeding crop exhibited maximum HUE in all varieties as because they produced maximum yield within minimum thermal use and had optimum temperature and sunlight and throughout growing period crop utilized heat more efficiently and increased biological activity that confirm higher yield (Fig. 2). These results are matched with the results reported by Chahal et al., 2007 and Jagtap *et al.,* 2018.

The second heat use efficient seeding date was found at 15 November seeding. The heat use efficiency decreased with delay seeding after 30 November seeding. BRRI dhan96 was found maximum heat use efficient variety (3.36 kg °C ha^{-1} days^{-1}) followed by BRRI dhan99 (2.95 kg °C ha^{-1} days^{-1}), BRRI dhan100 (2.78 kg °C ha^{-1} days^{-1}) and BRRI dhan97 (2.77 kg °C ha^{-1} days^{-1}).

Photothermal units

The accumulated photothermal units (PTU) followed the same trend as heat use efficiency (HUE) because both the indices depend on time period required for attaining physiological maturity. Here we observed higher PTU was between 15 and 30 December sowing because of longer growth duration and crop exposed to brighter sunshine. BRRI dhan96 and Bangabandhu100 had higher PTU at 15 November seeding (24,010 and 24,897 days °C hour respectively) whereas BRRI dhan97 and BRRI dhan99 had highest PTU at 30 December (2,682 and 26,079 days °C hour). Among all the seeding dates the lowest Photothermal units was observed at 30 November seeding (22146 to 24983 days °C hour). The highest grain yield at 30 Nov. seeding with minimum photothermal unit indicated that optimum environment prevailed for better growth and development for that seeding time.

Table 4. Response of yield to heliothermal units and heat use efficiency across different seeding dates and genotypes.

Variety		Seeding date			
		15th Nov	30th Nov	15th Dec	30th Dec
BRRI dhan96	Yield (kg/ha)	5900	6500	4600	3667
	HTU (days °C hour)	13607	12229	12618	12645
	HUE (Kg/ha/°C days)	2.79	3.36	2.39	1.91
BRRI dhan97	Yield (kg/ha)	5300	5867	5733	4300
	HTU (days °C hour)	14626	13670	13792	13845
	HUE (Kg/ha/°C days)	2.34	2.77	2.71	1.97
BRRI dhan99	Yield (kg/ha)	5267	6300	5167	4233
	HTU (days °C hour)	14626	13828	13792	13790
	HUE (Kg/ha/°C days)	2.32	2.95	2.46	1.95
BRRI dhan100	Yield (kg/ha)	5733	5767	4633	3200
	HTU (days °C hour)	13902	13307	13475	12870
	HUE (Kg/ha/°C days)	2.63	2.78	2.25	1.58

Table 5. Photothermal units (PTU) requirement of four varieties under different seeding dates.

Variety	Photothermal units (PTU) in days °C hour			
	15th Nov	30th Nov	15th Dec	30th Dec
BRRI dhan96	24010	22146	22392	22677
BRRI dhan97	25947	24442	24902	26282
BRRI dhan99	25947	24683	24651	26079
BRRI dhan100	24897	23909	24105	24197

Grain yield

Among the varieties, BRRI dhan96 produced statistically similar yield at 15 Nov. and 30 Nov. seeding (6.5 and 5.9 t/ha respectively). Similarly, there was no significant differences found in yield between 15 Nov and 30 Nov seeding for BRRI dhan97, BRRI dhan99 and BRRI dhan100, and the range of yield was 5.3 to 5.87 t/ha, 5.17 to 6.30 t/ha and 4.63 to 5.73 t/ha respectively (Fig. 1). The higher yield of 15 and 30 Nov seeding plant as it got maximum sunshine shine/solar radiation (Fig. 2) during their reproductive phase for photosynthetic activity. The average temperature was also optimum for crop growth (Fig. 2). This two sets plant also received maximum heat for a longer period of time which mean they got optimum temperature with lower minimum temperature at their vegetative phase which accumulate maximum food reservoir for grain development. It was found that if vegetative stage is too short to produce sufficient dry-matter the number of spikelet will be small (Vergara et al. 1964)

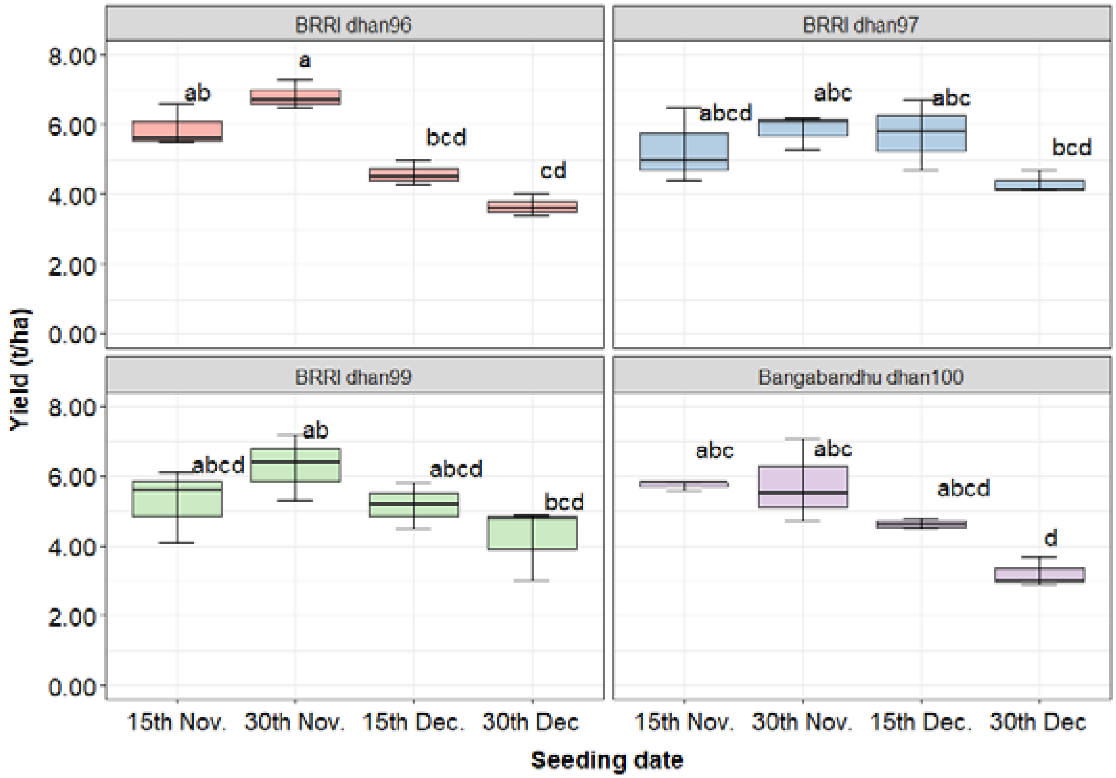
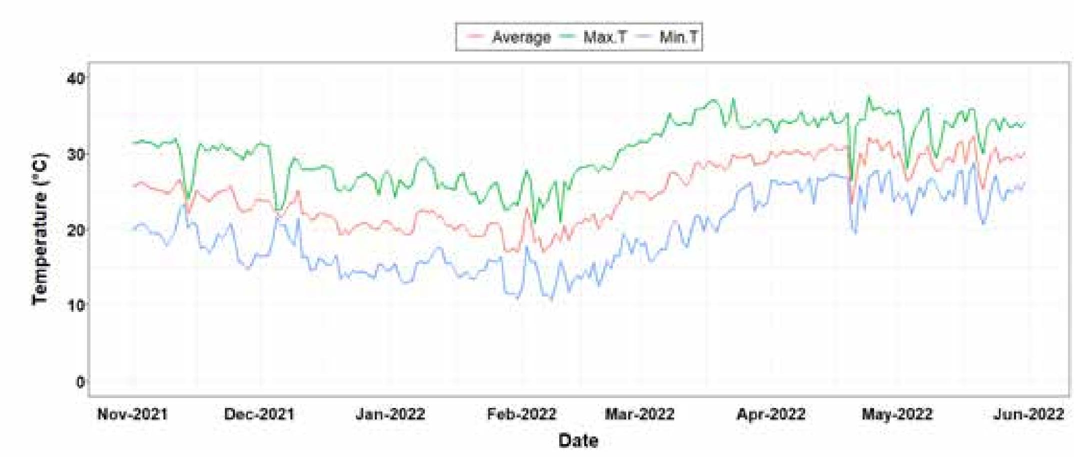


Fig. 1. Yield of four BRRI varieties under different seeding dates.



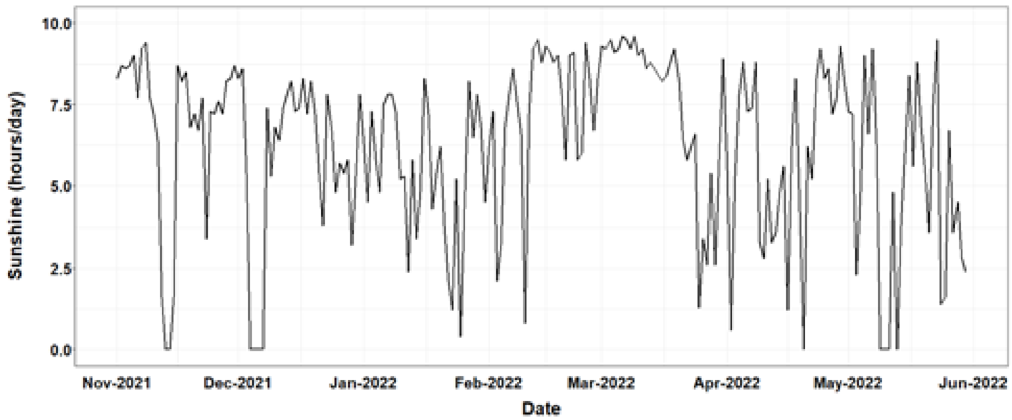


Fig. 2. Daily Maximum (Max.T), Minimum (Min.T), Average temperature (above graph) and daily sunshine hours during the crop growing period (below graph).

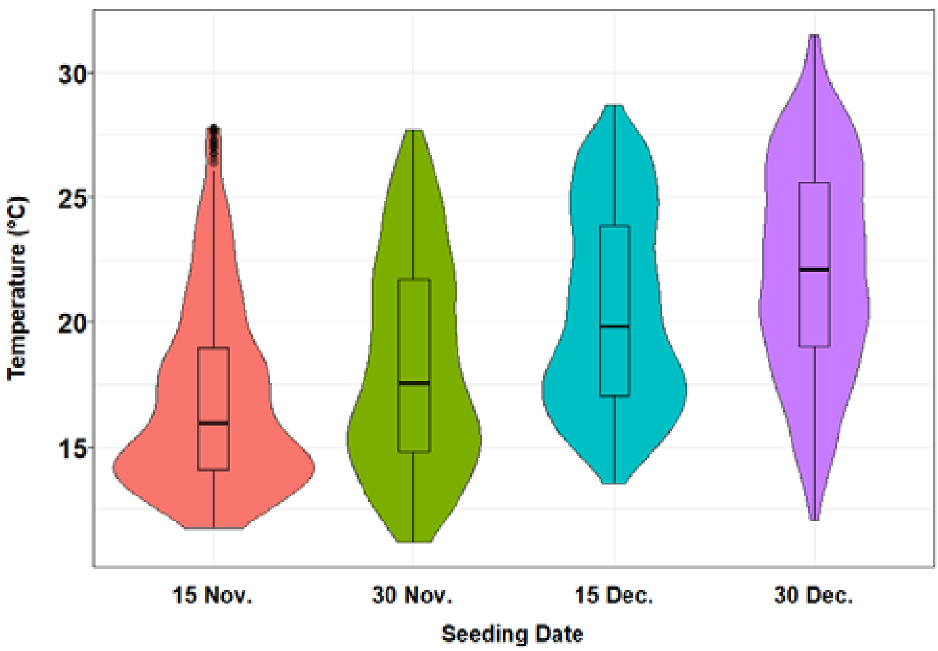


Fig. 3. The aggregated average temperature of every half an hour for 1st 15 days after transplanting of four different varieties. The data were taken from an automatic weather station (Davis Vantage Pro2 Weather Station) of the Plant Physiology Division, BRRI.

CONCLUSIONS

In conclusion, seeding in the last week of November resulted in higher heat use efficiency and produced a higher yield due to the crop experiencing optimal growing conditions. The total Growing Degree Days (GDD) required for physiological maturity was highest for the 15 November seeding and gradually decreased with later seeding dates up to the fourth seeding. It was observed that higher degree days up to the panicle initiation stage delayed crop maturity, while a decrease in degree days ultimately shortened the crop growth duration. Earlier seeding allowed the crop to accumulate more degree days, with each delay in transplanting reducing the degree day accumulation during the growing season. The Photothermal Units (PTU) were higher between 15 and 30 December sowing, attributed to the longer growth duration and increased exposure to brighter sunshine.

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Genetic Variability Analysis for Yield Contributing Traits in Rice (*Oryza sativa* L.) in Hilly Areas of Bangladesh

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ABSTRACT

A field experiment was carried out with six genotypes of Boro season rice at Sadar upazila of Khagrachhari district, Bangladesh. The study evaluated the genetic variability, heritability, correlations among the studied traits, and their direct effect on yield. The research result revealed that the phenotypic variance was higher than the genotypic variance for all the measured traits. High phenotypic coefficient of variation (PCV) and genotypic coefficient of variation (GCV) values were observed for grain yield (GY), thousand-grain weight (TGW), days to flowering (DTF), days to maturity (DTM), plant height (PH), panicle length (PL), number of filled grains per plant (FG), and number of unfilled grains per plant (UFG), indicating a very high heritability estimate. The calculated genetic advance as a percent of the mean for all traits was also very high. The GY was positively correlated with PH, DTM, and the number of total tillers per hill (NTH), PL, FG, UFG, and TGW; and negatively correlated with DTF and the number of non-effective tillers per hill (NETH). Principal component analysis revealed that PC1 exhibited the highest standard deviation (267.95) and proportion of variance (88%), with subsequent principal components showing gradual reduction. However, in terms of cumulative proportion, the contributions of the principal components were similar, except for PC1. Path analysis revealed that DTM, number of effective tillers per hill (ETH), PL, FG, UFG, and TGW directly affect GY. FG (0.526), PL (0.394), UFG (0.205), and ETH (0.192) showed a highly significant positive correlation contributed to the GY in path analysis; indicating that selecting these traits might be effective in improving grain yield in the future breeding programmes.

Key words: Genetic variability, correlation coefficients, path analysis, rice (*Oryza sativa* L.)

INTRODUCTION

Oryza sativa L., the most extensively farmed rice, is an indispensable food for an assessed 3.5 billion people globally and is the major crop in Asia, where around 90% of the globe's rice is growing and eaten up (Muthayya *et al.,* 2014 and Debsharma *et al.,* 2022). As a result of ever-rising inhabitants, the primary goal of plant breeders is to upturn rice production. However, several significant drivers have prevented the world from ending hunger and malnutrition in all its forms by 2030 (Byerlee and Fanzo, 2019).

Regarding global rice production, Bangladesh ranks third and is the fourth-largest rice user (Mottaleb *et al.,* 2020), with rice being the leading staple food. It is cultivated by 90% of the total net crop areas of the nation, and over 99% of the country's populace consumes 367.2 g of rice per person per day (HIES, 2016). Rice is grown in Bangladesh during three distinct seasons - Aus, Aman, and Boro, with Boro being the most important and leading season for rice in Bangladesh in terms of production volume in recent years. The Boro rice cultivation comprises about 61% of the total cropped area which is about 55% of the total rice

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production (BBS, 2019). Proper improved crop management practices such as balanced irrigation, fertilizer, and pesticide application have strengthened its production.

Rice cultivation in Khagrachari district, one of the three hill districts of Bangladesh, has great potential in the Boro season despite the less profitable traditional jhum cultivation. Specifically, the study aims to conduct trait-specific statistical varietal selection among the district's most cultivated inbred rice cultivars. The achievements of genetic variability by choice turn on the genetic progression of heredity, individual traits, and crop development, which rely on genetic diversity in the base population (Govintharaj *et al.,* 2016; Gupta et al., 2022). Once genetic variability is established, appropriate selection methods can improve crop yields by selecting yield components. Environmental factors play a significant role in the observable fluctuations of quantitative attributes. Therefore, partitioning overall variation is obligatory for a fruitful breeding platform. Heritability estimation helps predict the most effective selection strategy and breeding methods, as well as the profit from selection. A character with a high PCV and a low GCV has a high environmental effect on its manifestation (Singh, 2015). Although correlation estimates can reveal the positive or negative influence of factors on a trait, they are not capable of providing accurate insights into the relative significance of direct and indirect effects of intricate attributes like yield. Path analysis utilizes standard partial regression coefficients to differentiate the correlation coefficients between the direct and indirect effects of multiple attributes on the reliant variable. In this analysis, crop yield is regarded as the reliant variable, while residual characters are reflected as liberated variables.

Several researchers have used morphological traits for variability assessment and characterization of Bangladesh rice germplasm (Siddique et al., 2011; Banik et al., 2012; Khalequzzaman et al., 2012; Baktiar et al., 2013; Siddique et al., 2013; Islam et al., 2014; Ahmed et al., 2015a, 2015b; Kulsum et al., 2015; Akter et al., 2016; Biswash et al., 2016; Siddique et al., 2016; Akter et al., 2017; Islam et al., 2017; Akter et al., 2018; Islam et al., 2018; Siddique et al., 2018; Islam et al., 2019; Muti et al., 2020; Khalequzzaman et al., 2022; Khalequzzaman et al., 2023), but research exploring Boro rice is scanty. Thus, it is imperative to conduct this research with Boro rice to explore the genetic variation, correlation coefficients linked to characteristics that impact yield favourably or unfavourably, and the connection between yield components and their direct and indirect influences on grain yield via path coefficient analysis. As a result, this investigation was performed to assess the genetic variation, heritability, and correlation between diverse quantitative traits and their direct impact on yield via path analysis. This examination will establish the groundwork for rice selection and yield enhancement.

MATERIALS AND METHODS

An experiment was carried out during the Boro season of 2018-19 (December 2018 to May 2019) at a farmer's field located in Sadar upazila of Khagrachhari district, Bangladesh. The field belongs to the Northern and Eastern hills, which are categorized as the AEZ-29 (Agro-Ecological Zone) (Ahmmed *et al.,* 2018). The randomized complete block design (RCBD) was implemented in the trial, which comprised six inbred rice cultivars, specifically Binadhan-5, Binadhan-10, Binadhan-12, Binadhan-18, BRRI dhan28, and BRRI dhan29, with four replications. The experimental plot had a total area of 208 m^2, with individual plots measuring $2 \times 2\ m^2$. Pre-germinated seeds were sowed, and 2-3 seedlings were transplanted per hill at a spacing of 20 cm × 20 cm after 35 days of sowing. Fertilizer application involved administering urea, triple superphosphate (TSP), muriate of potash (MOP), gypsum, and zinc at the rate of 210-100-150-80-6 kg ha^{-1}. To control weeds, pre-emergence herbicide Rifit 500 EC @ 988 ml/ha was used, and two-hand weeding was done 30 and 45 days after transplanting. Pest infestation was prevented by applying Granular Carbofuran-5G @ 10 Kg/ha and spraying Amistar Top @ 500 ml/ha and Ripcord 10 EC @ /988 ml/ha. Harvesting was done when 80% of the grains matured and turned golden. Data were collected from five randomly selected competitive plants from each plot, including plant height (PH), number of total tillers per hill (NTH), number of effective tillers per hill (ETH), number of non-effective tillers per hill (NETH), panicle length (PL), number of filled grain per plant (FG), number of unfilled grain per plant (UFG), thousand grain-weight (TGW), days to maturity (DTM), days to flowering (DTF), and grain yield (GY) measured in ton per hectare.

Statistical Analysis

The Agricolae package of R statistical software in R Studio (Team R, 2017) was used for statistical analysis of various genetic parameters, including genetic variance, phenotypic variance, genotypic coefficient of variation (GCV), phenotypic coefficient of variation (PCV), heritability (h^2b), genetic advance (GA), and genetic advance as a percentage of the mean (GAM), as well as correlation coefficients and path analysis. Principal component analysis (PCA) was also performed using the GGally package in R Studio.

RESULTS

Performance of varieties

Recommendation of a variety for cultivation, considering the overall varietal performance, including grain yield and quality, is crucial for sustainable production. To evaluate the performance of six popular boro rice varieties, yield and growth characteristics were assessed. Variations in plant height (PH), days to flowering (DTF), days to maturity (DTM), total tillers per hill (NTH), effective tillers per hill (ETH), non-effective tillers per hill (NETH), panicle length (PL), filled grains per plant (FG), unfilled grains per plant (UFG), thousand grain-weight (TGW), grain yield (GY) were observed among the varieties. Analyzing the ANOVA (Table 1) and mean performance (Table 2) of different yield-related traits of six varieties of the best-performing varieties were identified.

Table 1. Analysis of variance (mean sum of squares) for yield and its component characters of six popular Boro rice varieties.

Source of variation	df	PH (cm)	DTF	DTM	NTH	ETH	NETH	PL (cm)	FG (No.)	UFG (No.)	TGW (g)	GY (t/ha)
Varieties	5	142.64***	233.76***	384.38***	3.58***	6.76***	16.15***	21.54***	27.80***	25.37***	851.58***	31.24***
Replication	3	1.24	10.20***	9.13***	2.83*	2.45	2.06	1.76	2.36	2.08*	9.02***	5.33**
Error	15	4.14***	*	*	2.03*	1.70	1.02	0.97	3.86***	2.85***	2.75***	*

*, ** and *** indicates significant at 0.05, 0.01 and 0.001 probability, respectively.

PH: plant height, NTH: number of total tillers per hill, ETH: number of effective tillers per hill, NETH: number of non-effective tillers per hill, PL: panicle length, FG: number of filled grains per plant, UFG: number of unfilled grains per plant, TGW: thousand grain-weight, DTM: days to maturity, DTF: days to flowering and GY: grain yield.

Table 2. Mean performance of six varieties based on different morphological traits related to yield.

Varieties	PH (cm)	DTF	DTM	NTH (no.)	ETH (no.)	NETH (no.)	PL (cm)	FG (no.)	UFG (no.)
Binadhan-5	115.20 a	127.50 c	161.00 b	11.05 b	10.25 b	0.80 cd	23.58 ab	888.13 bc	347.00 ab
Binadhan-10	107.25 b	106.25 e	139.00 d	12.65 ab	11.50 b	1.15 bcd	22.96 ab	835.74 cd	180.83 d
Binadhan-12	94.70 c	126.00 c	159.00 b	14.30 a	13.85 a	0.45 d	24.35 a'	1262.75 a	379.75 a
Binadhan-18	93.50 cd	130.75 b	151.00 c	14.10 a	10.00 b	4.10 a	20.12 d	722.50 d	258.50 c
BRRI dhan28	92.25 cd	119.75 d	143.75 c	13.60 ab	11.65 ab	1.95 bc	20.90cd	693.75 d	160.37 d
BRRI dhan29	91.25 d	134.25 a	166.50 a	12.75 ab	10.45 b	2.30 b	22.27 bc	1001.50 b	288.75 bc
P value									
Varieties	0.000	0.000	0.000	0.135	0.012	0.000	0.000	0.000	0.000
Replication	0.738	0.288	0.441	0.233	0.236	0.154	0.188	0.490	0.295
CV (%)	9.82	6.72	7.65	23.32	24.59	106.32	9.51	29.05	41.66
SEM	0.89	0.87	0.95	0.28	0.25	0.17	0.19	23.9	10.2
Mean	99.03	124.08	154.88	13.08	11.28	1.79	22.36	900.70	269.20

The same letter indicates no significant differences and the different letter indicates significant differences among the varieties for each trait. PH: plant height, NTH: number of total tillers per hill, ETH: number of effective tillers per hill, NETH: number of non-effective tillers per hill, PL: panicle length, FG: number of filled grains per plant, UFG: number of unfilled grains per plant, TGW: thousand grain-weight, DTM: days to maturity, DTF: days to flowering and GY: grain yield. CV: Coefficient of variation, SEM: Standard error mean.

Estimation of Genetic Variability

Improvement of field crops (like rice) through breeding, the level of diversity for each given trait is necessary. Table 3 presents the assessments of genotypic variation (σ^2g), phenotypic variation (σ^2p), error variance (σ^2e), GCV, PCV, heritability, GA, and GAM for the previously mentioned eleven traits. Using only genotypic variance as a comparison to total phenotypic variance, we determined the extent of the role of genotype in rice varietal development.

For all traits considered, genotypic variance exceeded environmental variance at the mean level excluding the NTH of σ^2g=5.20 and σ^2p=13.25. In the present study, GCV value varied from 14.00% for DTM to 160.43% for the NETH, whereas PCV varied from 14.12% for DTM to 180.37% for the NETH. The greater PCV and GCV values were recorded for GY, PH, ETH, NETH, FG, UFG, and TGW where not extreme GCV and PCV values had been obtained for DTM, DTF, and PL. Among the studied traits, TGW, DTM, DTF, PH, PL, GY, FG, and UFG revealed more than 80% of heritability estimates.

Table 3. Assessment of genetic factors for GY and yield attributes.

Character	σ^2g	σ^2p	σ^2e	CV		h^2b	GA	GAM (%)
				GCV (%)	PCV (%)			
DTF	484.92	493.25	8.33	17.76	17.91	98.31	44.97	36.27
DTM	470.72	479.04	8.31	14.00	14.12	98.26	44.30	28.59
PH	481.46	495.06	13.60	22.16	22.47	97.25	44.58	45.02
NTH	5.20	13.25	8.05	17.44	27.84	39.25	2.94	22.51
ETH	8.67	14.69	6.02	26.10	33.97	59.02	4.66	41.30
NETH	8.26	10.44	2.18	160.43	180.37	79.11	5.27	293.94
PL	12.35	14.75	2.40	15.71	17.17	83.70	6.62	29.61
FG	212281.28	243964.66	31683.38	51.15	54.84	87.01	885.35	98.29
UFG	36908.63	42966.00	6057.37	71.37	77.00	85.90	366.80	136.26
TGW	95.86	96.31	0.45	43.65	43.75	99.53	20.12	89.71
GY	1.73	1.96	0.23	21.43	22.81	88.32	2.54	41.50

σ^2g: Genotypic variance, σ^2p: Phenotypic variance, σ^2e: Environmental variance, GCV: Genotypic coefficient of variation, PCV: Phenotypic coefficient of variation, h^2b: broad sense heritability, GA: Genetic advance and GAM: GA as a percent of the mean.

Here, PH: plant height, NTH: number of total tillers per hill, ETH: number of effective tillers per hill, NETH: number of non-effective tillers per hill, PL: panicle length, FG: number of filled grains per plant, UFG: number of unfilled grains per plant, TGW: thousand grain-weight, DTM: days to maturity, DTF: days to flowering and GY: grain yield.

Likewise, moderately high heritability (60-79%) was documented for the NETH. Instead, a moderate broad sense heritability estimate (40-59%) was perceived for the ETH and less H^2 (<39%) was shown for the NTH. The estimated GA as a GAM in the current study was higher and it was about >20% for all the traits. The uppermost value of GAM (293.94) was detected in the NETH, tracked by 136.26 in the UFG, 98.29 in the FG, 89.71 in TGW, and 45.02 in PH.

Principal component analysis (PCA)

For summarizing the information of large data, principal component analysis effectively decreases the measurement dimensions while retaining data accuracy. In principal component analysis, the largest standard deviation and proportion of variance were found in PC1 (267.95 and 88%, respectively) and for others PC was gradually reduced but in the case of cumulative proportion, the PCs were similar except PC1 (Table 4).

In PC1, the variable FG has a high negative coefficient (-0.97) and UFG has a low negative coefficient (0.23). This suggests that PC1 is capturing information related to the contrast between filled and unfilled grains. PC2 has notable positive coefficients for DTF, DTM, and UFG, suggesting that PC2 may be associated with the timing of growth stages and unfilled grains. PC3 has a high positive coefficient for PH, ETH, PL, UFG, TGW, and GY indicating that PC3 may represent variations related to plant height, effective tiller per hill, panicle length, unfilled grain per plant, thousand-grain weight, and grain yield. On the other hand, PC3 has a high negative coefficient to DTM and DTF. Again, PC4 has a high negative coefficient to PH, DTM, and DTF which represents variability. Similarly, the other principal components (PC5 to PC11) capture specific patterns or relationships among the variables (Table 5).

The PCA results provide a way to understand the relationships and patterns in the data, reducing the dimensionality of the original variables while retaining important information. The coefficients in each principal component indicate the contribution of each variable to the overall variation captured by that component.

Table 4. Standard deviation, proportion of variance and cumulative proportion of each principal components for the principal components of six rice varieties.

Parammeter	PC1	PC2	PC3	PC4	PC5	PC6	PC7	PC8	PC9	PC10	PC11
Standard deviation	267.95	96.23	13.50	8.31	3.69	3.35	1.81	1.60	1.25	0.50	0.00
Proportion of variance	0.88	0.11	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Cumulative proportion	0.88	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00

Table 5. Principal component analysis for quantitative traits of six rice varieties.

Variable	PC1	PC2	PC3	PC4	PC5	PC6	PC7	PC8	PC9	PC10	PC11
PH (cm)	0.00	0.01	0.48	-0.85	0.14	0.06	0.10	-0.05	-0.06	0.03	0.00
DTM	0.00	0.04	-0.58	-0.43	-0.03	-0.12	-0.30	0.18	0.57	-0.14	0.00
NTH	0.00	0.00	0.00	0.10	0.73	-0.27	0.10	0.19	0.05	-0.01	-0.58
ETH	0.00	0.00	0.05	0.08	0.59	0.02	-0.52	-0.22	-0.05	0.00	0.58
NETH	0.00	0.00	-0.04	0.02	0.15	-0.30	0.61	0.41	0.10	0.00	0.58
DTF	0.00	0.03	-0.64	-0.26	0.13	0.06	0.19	-0.18	-0.64	0.15	0.00
PL (cm)	0.00	0.00	0.06	-0.03	-0.06	0.20	-0.36	0.82	-0.39	-0.06	0.00
FG	-0.97	-0.23	-0.01	-0.01	0.00	0.00	0.00	0.00	0.00	0.00	0.00
UFG	-0.23	0.97	0.04	0.04	0.00	-0.01	0.00	0.00	0.00	0.00	0.00
TGW	0.01	-0.01	0.10	-0.06	-0.25	-0.88	-0.27	-0.07	-0.27	-0.03	0.00
GY (t/ha)	0.00	0.00	0.01	0.00	-0.03	-0.05	-0.10	0.11	0.15	0.98	0.00

Here, PH: plant height, NTH: number of total tillers per hill, ETH: number of effective tillers per hill, NETH: number of non-effective tillers per hill, PL: panicle length, FG: number of filled grains per plant, UFG: number of unfilled grains per plant, TGW: thousand grain-weight, DTM: days to maturity, DTF: days to flowering and GY: grain yield.

Coefficients of Correlation Analysis

The association among the physiological and yield-attributing traits was calculated through a correlation study concerning them (Table 6). The GY was positively correlated with PH, NTH, ETH, PL, FG, UFG, TGW, DTM and negatively correlated with DTF and NETH. Among the traits, GY has a positive significant correlation with ETH, PL, FG, and UFG and simultaneously, a negative significant correlation with DTF and NETH.

Table 6. Coefficients of correlations among diverse yield attributing characters of six rice genotypes.

Traits	PH	DTF	DTM	NTH	ETH	NETH	PL	FG	UFG	TGW
DTF	-0.321***									
DTM	-0.126	0.918***								
NTH	-0.162	-0.001	-0.037							
ETH	0.001	-0.156	-0.121	0.790***						
NETH	-0.261**	0.226*	0.118	0.450***	-0.192*					
PL	0.332***	-0.127	0.042	-0.115	0.170	-0.431***				
FG	-0.021	0.155	0.312***	0.189*	0.392***	-0.269**	0.435***			
UFG	0.106	0.441***	0.519***	-0.008	0.132	-0.205*	0.389***	0.480***		
TGW	0.251**	-0.438***	-0.391***	-0.084	-0.275**	0.266**	-0.289***	-0.478***	-0.444***	
GY	0.091	-0.205*	0.006	0.055	0.192*	-0.191*	0.394***	0.526***	0.205*	0.011

Path Coefficient Analysis of the Concerned Traits

Determining the direct and indirect effects allowing for ten characters viz. PH, DTM, NTH, ETH, NETH, DTF, PL, FG, UFG, and TGW, the path coefficient analysis was performed utilizing a correlation coefficient. The traits PH, DTM, NTH, ETH, PL, FG, UFG, and TGW had positive direct effects on GY (Table 7). The high values of direct positive effects of FG (0.526) displayed a highly significant positive correlation and this trait contributed the maximum for GY. Besides, the direct effects of other considered characters on grain yield were optimistic, resilient, and significant for the characters like PL (0.394), UFG (0.205), and ETH (0.192). In contrast, a negative direct influence on grain yield per plant was exercised by NETH (-0.191) and DTF (-0.205).

Table 7. Partitioning of phenotypic correlations into direct and indirect influences of ten important traits by path analysis.

Traits	PH	DTM	NTH	ETH	NETH	DTF	PL	FG	UFG	TGW	Correlation to GY
PH	-0.382	-0.481	0.157	0.000	-0.170	0.892	0.046	-0.006	0.003	0.031	0.091
DTM	0.092	**2.005**	0.137	-0.268	0.131	-2.196	-0.021	0.009	0.009	-0.037	0.006
NTH	0.061	-0.281	**-0.979**	0.729	0.294	0.206	-0.015	0.056	0.000	-0.010	0.055
ETH	0.000	-0.581	-0.774	**0.923**	-0.124	0.641	0.024	0.114	0.003	-0.035	0.192*
NETH	0.099	0.401	-0.441	-0.175	**0.654**	-0.618	-0.059	-0.079	-0.005	0.034	-0.191*
DTF	0.149	1.924	0.088	-0.258	0.176	**-2.288**	-0.036	-0.020	0.007	-0.042	-0.205*
PL	-0.126	-0.301	0.108	0.157	-0.281	0.595	**0.138**	0.126	0.010	-0.036	0.394***
NFG	0.008	0.060	-0.186	0.360	-0.176	0.160	0.059	**0.292**	0.013	-0.060	0.526***
NUFG	-0.042	0.682	0.010	0.120	-0.131	-0.595	0.054	0.140	**0.027**	-0.055	0.205*
TGW	-0.095	-0.601	0.078	-0.258	0.176	0.778	-0.040	-0.140	-0.012	**0.125**	0.011
Residual Effect = 0.421											

*, **, and *** indicate significance at 5%, 1%, and 0.1% levels, respectively. Bold figures indicate the direct influence.

DISCUSSION

Success in crop breeding programmes is largely determined by genetic variability and the transmission of desired traits. Exploring the genetic variability, and trait associations of a species with the intervention of plant breeders may result in the improvement of desired varieties. The grain yield is a complicated product affected by many interdependent quantitative characters. Plant breeders have a blueprint for selection if they understand how other traits affect yield and how genetic and non-genetic components interact.

Explanation of quantitative traits for crop improvement

The assessment of genotypic and environmental variances provided valuable insights into the underlying factors influencing the observed trait variations. Notably, the genotypic variance exceeded environmental variance at the mean level, excluding the NTH with $\sigma^2g=5.20$ and $\sigma^2p=8.05$. This suggests a predominant genetic influence on the traits under investigation, highlighting the significance of genetic factors in determining phenotypic expressions. The GCV and PCV values further elucidated the variability within the studied traits. Substantial variations were observed, with GCV ranging from 14.00% for DTM to 160.43% for NETH, and PCV ranging from 14.12% for DTM to 180.37% for NETH. Notably, traits such as GY, PH, ETH, NETH, FG, UFG, and TGW exhibited higher PCV and GCV values, indicating a greater potential for genetic improvement in these traits. Conversely, DTM, DTF, and PL displayed lower GCV and PCV values, suggesting a comparatively lower genetic variability in these traits. The outcomes described by Sravan *et al.* (2012) and Karim *et al.* (2007) comply with the very low alteration between PCV and GCV, which indicates that the environment has little influence on the manifestation of the trait or that genotypes are less sensitive to the environment. The findings from earlier studies (Bitew, 2016; Hossain *et al.,* 2015) are consistent with the findings of our investigation, demonstrating that ecological influence is not discernible in the expression of phenotypic traits. As a result, features with lesser fluctuation by the environment and choice attributed to the phenotype reasonably than the genotype may be suitable for improving such potentials (Karad and Pol, 2008). Studies have also shown that on the part of generating high-yielding cultivars over and done with hybridization and selection, cultivars with high GCV of yield-contributing traits are essential. The characters' having high and low PCV with GCV differences reveal their vulnerability to environmental change and their differential influence with it. The PCV and GCV values of more than 20% are high, less than 10% are low, and 10% to 20% are considered medium, according to Girma *et al.* (2018). According to Bose *et al.* (2007), a high level of GCV results in varied offspring in the segregating generations. For the parameters, FG, test weight, and yield, the degree of PCV and GCV were moderate to high (Roy *et al.,* 2001; Thirumala *et al.,* 2014). For FG, test weight and yield. Thirumala *et al.* (2014) once more reported high PCV. The PCV and GCV were moderate to high (Lingaiah *et al.,* 2014).

The heritability estimates provided further insights into the genetic control of the traits, with TGW, DTM, DTF, PH, PL, GY, FG, and UFG demonstrated heritability estimates exceeding 80%. NETH showed moderately high heritability (60-79%), while ETH exhibited a moderate broad-sense heritability estimate (40-59%), and NTH showed low heritability (<39%). These findings underscore the potential for successful selection for breeding programs, particularly for traits with high heritability. In this study, the poor heritability of NTH (39.25%) was attributed to the substantial environmental influence on this character expression. Genetic advance as a percentage of the mean values indicated substantial genetic variability, with NETH exhibiting the highest GAM (293.94), followed by UFG, FG, TGW, and PH. These values suggest ample scope for genetic improvement through selection. The heritability assessments besides GA can be suitable for calculating the effect of selection in a breeding programmes. Vaithiyalingan and Nadarajan (2006) stated that the traits viz. NGP (number of grains per plant), test weight, yields, and PH revealed a high level of GA as a percent of the mean. Lingaiah *et al.* (2014) stated that high heritability united with high GAM for the traits' number of grains per panicle, TGW, and yield representing the role of the additive genes in leading these characters. The PH, seeds per plant, and number of spikelets per panicle demonstrated high heritability in combination with high to moderate GAM, representing the preponderance of additive gene action in the development of these features (Yadav *et al.,* 2011). According to the current study's findings of higher heritability and high GAM, selection would be successful because of additive gene action in the expression of character (Hossain *et al.,* 2015).

Therefore, the comprehensive analysis of genotypic and environmental variances, along with GCV, PCV, heritability, and genetic advance values, provides a robust foundation for understanding the genetic basis of the studied traits. The identified traits with high heritability and substantial genetic variability offer promising avenues for targeted breeding efforts aimed at enhancing rice cultivars.

Principal component analysis (PCA)

For six varieties 11 principal components emerged as a robust method for summarizing information from a large dataset for explaining the varietal traits where the cumulative proportion of variation ranged from 88% to 100%. However, cumulative variance with maximum variability for three major PCs was stated by Basavaraj *et al.* (2022). Neeru *et al.* (2016) also acknowledged 11 principal components (PCs) which explained about 75% variability in mustard.

The analysis revealed that PC1 played a prominent role, exhibiting the largest standard deviation (267.95) and accounting for 88% of the total variance (Table 4). Rahangdale *et al.* (2021) and Debsharma *et al.* (2024) reported a maximum variation percentage in PC1 in their study on 67 rice lines. Similar findings were reported by Shivani *et al.* (2021), Manohara *et al.* (2020), and Sahu *et al.* (2016).

This signifies the dominance of PC1 in capturing the overall variability in the dataset (Table 4). Examining the loadings within PC1 provided valuable insights into the information encapsulated by this principal component. The variable FG displayed a high negative coefficient (-0.97), while UFG had a low negative coefficient (0.23); suggesting that PC1 primarily captures information related to the contrast between filled and unfilled grains. This aligns with the notion that PC1 serves as a key indicator of grain characteristics. Moving to PC2, the presence of notable positive coefficients for DTF, DTM, and UFG implies an association with the timing of growth stages and the occurrence of unfilled grains. This suggests that PC2 may provide insights into the temporal aspects of plant development, particularly regarding grain filling. The subsequent principal components (PC3 to PC11) further contributed to the understanding of specific patterns or relationships among the variables (Table 5). Similar findings were observed by Kishore et al. (2007), and Babar et al. (2009). Using path analysis, the eight traits were estimated to contribute 58 percent of grain yield variability where the residual influence was 0.42.

Coefficients of correlation for the studied associated traits

Positive correlations were observed between GY and PH, NTH, ETH, PL, FG, UFG, TGW, and DTM; while negative correlations were noted with DTF and NETH. Specifically, GY exhibited significant positive correlations with ETH, PL, FG, and UFG, concurrently displayed significant negative correlations with DTF and NETH among the studied traits. These findings highlight the interconnectedness of physiological attributes and yield-related characteristics, providing valuable insights into the complex relationships that influence overall grain yield. The positive associations suggest that enhancements in certain traits, such as ETH, PL, FG, and UFG, may contribute positively to grain yield, while negative correlations with DTF and NETH indicate potential trade-offs that need consideration in breeding or management strategies (Table 6). In the correlation coefficient among the grain yield per plant and additional measurable characters contributing to yield, Yadav *et al.* (2011) found that the GY was significantly and positively related to the NTH, PH, the number of panicles per plant, the number of spikelets per panicle and TGW at in cooperation to genotypic and phenotypic levels. According to Debsharma et al. (2020) and Eidikohnaki et al. (2013), there is a significant positive association between ETH and GY. The positive connection of GY with filled grains per panicle was reported by Perween *et al.* (2020) and Eidikohnaki *et al.* (2013). These mentioned research results are similar to the current study.

Path coefficient analysis of the studied associated traits

Plant breeders have utilized path analysis in agriculture to help them uncover features to increase crop yield which is the most important trait for the crop breeders (Milligan *et al.,* 1990). In this analysis, the direct and indirect stimuli on grain yield were examined. Based on these studied associated traits, the selection efficiency might be improved where it expedites the selection process and save time and money. By exploiting variable cultivars and retaining effective selection practices to enhance yield through yield traits, breeding initiatives attempt to raise rice production. Correlation analysis is a crucial step in determining the association between the yield and yield-contributing traits but path analysis, which separates a trait's influence into direct and indirect influences, is even more vital for the choice of yield-contributing traits (Dhavaleshvar *et al.,* 2019). According to Yadav *et al.* (2011), NTH, PL, NSP, and PH are the primary factors that affect yield since they have a direct beneficial impact on seed production per hill. According to Satheeshkumar and Saravanan (2012), path analysis demonstrated the greatest direct benefits for kernel length, FG, total grains produced, and ETH. Positive direct influences and correlation coefficients suggested that choice for these characters could be used to increase yield. Makwana *et al.* (2010) observed similar findings. The current study found that DTM, ETH, PL, FG, UFG, and TGW traits should be considered in choosing the genotypes for increased GY of rice is crucial. The top-most significant characters that directly contributed to GY are the DTM, ETH, PL, FG, UFG, and TGW, according to the findings of correlation and path analysis.

CONCLUSION

The studied genotypes possess promising characteristics and have the potential to be integrated into the breeding of diversified rice cultivars for the hilly ecosystem of Bangladesh. The PCV was observed to be marginally greater than the GCV indicating the minimum influence of the environment on the expression of the traits. The existence of additive genetic mechanisms and negligible environmental effects, as evidenced by the high heritability combined with a high GAM and genotypic correlation coefficients that were superior to their phenotypic correlation coefficients, further enhances inherent genetic associations. Therefore, it is advisable to choose these genetic parameters as a high priority in selection for further development, which could be utilized in future rice breeding programmes for the hilly areas.

AUTHORS' CONTRIBUTIONS

R.G.: Design, formulation, and supervision of experiment, writing of manuscript, performing the field and collection and analysis of data. S.K.D.: Analysis of data and writing of the manuscript. N.J.: writing of the manuscript. M.H. R.: supervision of experiment, and review of manuscript.

DECLARATION OF INTERESTS

The authors have declared that no competing interest exists.

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Operational Efficiency of Combine Harvesters on Different Field Size in the Selected Area of Bangladesh

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ABSTRACT

The length and width of the rice field play a crucial role in the performance of the combine harvester. Effect of field size (length of the field) on field performance of head feed (Kubota PRO588I-G and Yanmar AG600A) and whole feed (Yanmar YH700 and FM World WM 4LZ-4.0EA) combine harvesters were assessed in both irrigated dry season 2021-22 and non-irrigated wet season 2022 in two different regions of Bangladesh. Five levels of field length, i.e. ≤30m (L1), 30-40m (L2), 41-50m (L3), 51-60m (L4), and 61-70m (L5), were chosen to investigate forward speed, theoretical field capacity, effective field capacity, and field efficiency. The results revealed that all the performance parameters increased significantly with the increase of field length for all types of combine harvesters in both seasons. In the irrigated dry season, forward speed varied from 1.4 to 3.4 and 4.0 to 6.7 km/h of the head feed and whole feed combine harvester respectively with the field length L1 to L5, whereas significantly higher forward speed was observed for whole feed combine harvesters. On contrary, the field efficiency varied from 20 to 64 and 23 to 65% of the head feed and whole feed combine harvesters respectively with the field length L1 to L5. In the non-irrigated wet season, forward speed varied from 3.1 to 4.5 and 3.7 to 4.8 km/h of the head feed and whole feed combine harvesters respectively with the field length L1 to L5. Contrary, the field efficiency varied from 40 to 80 and 38 to 76% of the head feed and whole feed combine harvesters respectively with the field length L1 to L5. The field length for the studied combine harvester should not be less than 41-50 m to obtain more than 50% field efficiency of the machine in both the irrigated dry and non-irrigated wet season in Bangladesh.

Key words: Combine harvester, forward speed, actual field capacity, field efficiency, rice harvest

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INTRODUCTION

Bangladesh's rice sector has made significant progress in mechanization as a result of several governmental and non-governmental activities over the past few years, specially focused on paddy harvesting. The upshot was that Bangladesh's agricultural economy became one of South Asia's most mechanized (Islam and Shirazul, 2009 and Baudron *et al.,* 2015). Harvesting nourmally accounts for 24.9% of the total labor requirement for rice cultivation, which is more than other rice production activities. Total labour requirement of rice production in Bangladesh is about 149 man-hr/ha (Ali et al., 2019). In 2016, mechanized harvesting of paddy made up about 2% (MoA, 2016); today, it makes up 18% (Hossen, 2023). The Bangladesh government is implementing a project of Taka 30.2 billion entitle "Farm Mechanization through Integrated Management" to distribute 51,300 units of agro-machinery (combine harvester: 15,000 units) from 12 categories during 2020-2025 giving special importance on paddy harvesting and transplanting through the Department of Agricultural Extension (Financial Express, 2022). A total of 7,256 combine harvester already distributed to farmers under the above project (Bangladesh Post, 2023). Hence, combine harvesters are becoming more and more popular as an alternative to the traditional methods of harvesting and threshing of rice. Both head feed and whole feed combine harvesters are available in Bangladesh with different size and specifications. Medium to large type combine harvester, horse power rages 50-120, is importing under this project (Financial Express, 2022) while the average farm size in the country has decreased to less than 0.6 hectare, and 58 percent of people lack access to land (Financial Express, 2021). Normally large field are pre-requisite for efficient operation of the combine harvester. Other factors such as field size and shape, soil condition, crop condition, load bearing capacity of soil., etc. influence the field performance of the both head feed and whole feed combine harvester (Islam *et al.,*2020). This study has been conducted to identify the effect of field size on the performance of the both head feed and whole feed combine harvesters available in Bangladesh that will help the policy maker to estimate the suitable rice area and number of combine harvester required for sustainable mechanization. In addition, it would help the users to operate the combine harvester in profitable way

MATERIALS AND METHODS

This study has been conducted at Sadar Upazila of Habiganj district (24.351263 N, 91.424143 E) and Raiganj upazial of Sirajganj districts (24.5295° N, 89.5452° E) of Bangladesh during the irrigated dry season (Boro season) of 2021-22 and non-irrigated wet season (Aman season) in 2022 (Fig. 1). Both the head feed and whole feed combine harvesters were studied. Kubota PRO588I-G (Head feed) and Yanmar YH700 (Whole feed), two popular models available in the study areas, were used in Boro season, 2021-22 while FM World WM 4LZ-4.0EA (Whole feed) and Yanmar AG600A (Head feed) model combine harvesters were used in Aman 2022 season.

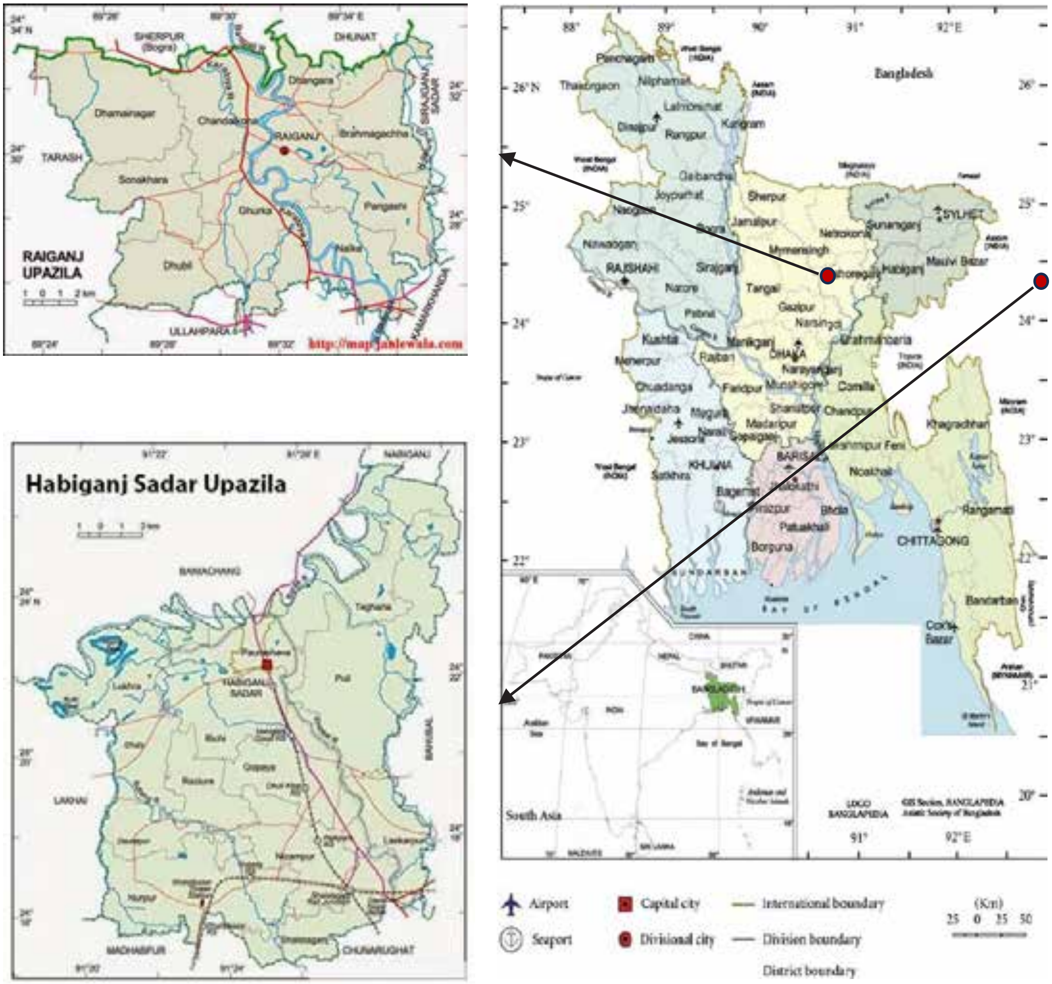


Fig. 1. Location of the study.

Experimental design

This study has been conducted in two different locations of the country and two different seasons (Table 1). Both the head feed and whole feed type combine harvesters used to determine the effect of field length on the performance of the machine. Hard soil layer of the field in the respective locations was almost same, which was measured manually during machine operation (Table 1). Soil type and field conditions during machine operations are presented in the Table 1.

Table 1. Experimental design and field conditions.

Season	Location	Model	Area (ha)	Length of the field (m)	Width of the field (m)	Depth of hard soil layer (mm)	Soil type	Field status during operation
Boro, 2021-22	Sirajganj	Kubota PRO588I-G	0.3515	62	57	2.5	Sandy	wet and 10.5 mm standing water
		Yanmar YH700	0.2015	63	32	2.7	Sandy	wet and 10.75 mm standing water
	Habiganj	Kubota PRO588I-G	0.224	65	34	38.1	Sandy loam	wet and 11.5 mm standing water
		Yanmar YH700	0.4905	75	65	38.5	Sandy loam	wet and 12.0 mm standing water
Aman, 2022	Sirajganj	Yanmar AG600A	0.456	70	65	17.1	Sandy	dry
		FM World Ruilong	0.452	71	64	18.5	Sandy	dry
	Habiganj	Yanmar AG600A	0.454	73	62	24.0	Sandy loam	dry
		FM World Ruilong	0.426	67	64	23.5	Sandy loam	dry

Note: Three trials were conducted in each site of the respective model of combine harvester.

Crop condition and yield

In Boro 2021-22 season, BRRI dhan89 and BRRI dhan29 were harvested in the locations of Sirajgaj and Habiganj while BRRI dhan49 and BRRI dhan75 were harvested in Aman 2022 season (Table 2). Prior to operation, plant height, grain moisture content, grain yield, grain maturity during harvesting, and cutting height from the ground after harvesting of the crops were measured (Table 2).

Table 2. Crops attributes and yield of the experimental field.

Season	Location	Paddy variety	Plant height (mm)	Grain moisture content (%)	Grain maturity during harvesting (%)	Cutting height from the ground (mm)	Grain yield (t/ha) at 14% MC
Boro 2021-22	Sirajganj	BRRI dhan89	1163	24.61	85	196.2	7.15
		BRRI dhan89	1194	25.66	85	345	7.34
	Habiganj	BRRI dhan29	932	26.84	90	195.5	6.34
		BRRI dhan29	951	27.69	90	348	6.31
Aman, 2022	Sirajganj	BRRI dhan49	1146	23.06	90	75	5.47
		BRRI dhan49	1122	22.79	90	250	4.82
	Habiganj	BRRI dhan75	1032	22.13	85	75	6.14
		BRRI dhan75	1038	22.29	85	250	5.39

Physical parameters of the combine harvester

Prior to field study, dimensions, load bearing capacity, and cutting width were measured; nevertheless, general data of each model of the combine harvesters and engine were also recorded from the manufacturers' specification (Table 3).

Field performance test

Field performance of the studied combine harvesters was tested in two different locations during the Boro 2021-22 and Aman 2022 season. Field performance was determined using various field lengths ≤30m, 31-40m, 41-50m, 51-60m, and 61-70m. In each case, the main length of the field was divided into the appropriate length type for the investigation. The lengthwise time per pass, without accounting for turning or any other losses, was measured in order to determine the theoretical field capacity of the machine as well as its forward speed. Total operational time and total area were measured to calculate the effective field capacity for different field length. The machine's field efficiency in the specified type of field was calculated using both the actual and theoretical field capacity.

Forward speed was determined by dividing the distance by the time needed to run the machine over that distance. The forward speed of combine harvester is determined using the following equation (Hunt,1995).

$$S = \frac{D}{T} \times 3.6$$

Where,

S= Forward speed of the machine, km/hr

D= Distance covered by the combine harvester, m

T= Time required to cover that distance, sec

Table 3. Physical parameters of the studied combine harvesters

Item	Model			
	Kubota PRO588I-G	Yanmar YH700	Yanmar AG600A	WM 4LZ-4.0EA
1. General Information				
1.1: Brand	Kubota	YANMAR	YANMAR	FM WORLD
1.2: Country of Manufacturer	China	China	China	CHINA
1.3: Country of origin	Japan	Japan	Japan	CHINA
1.4: Types	Head Feed	Whole Feed	Head Feed	Whole Feed
2. Dimensions				
2.1: Overall length × width × height (mm)	4240×1900×2800	5070×2285×2820	2990×1940×2410	49600×2890×2700
3. Engine				
3.1: Overall weight (kg)	2705	3571	3117	3200
3,2: Displacement (CC)	2434	3318	3318	3300
3.3: Engine power (kW)	49.2	51.5	47.59	65.65
3.4: Fuel tank capacity (l)	50	115	67	150
3.5: Oil tank Capacity (L)	9.1	9.4	9.4	9.0
4. Machine and travelling				
4.1: Grain tank Capacity (kg)	600	1500	1000	1200
4.2: Steering	HST	HST	HST	HST
4.3: Gearshift	Manual (3 steps)	Manual (3 steps)	Manual (3 steps)	Manual (3 steps)
4.4: Forward speeds	0 to 2.05 max	0 to 3.00 max	0 to 1.65 max	0 to 2.56 max
4.5: Reverse speeds	0 to 2.05 max	0 to 3.00 max	0 to 1.65 max	0 to 2.56 max
4.6: Driving wheel/crawler	Crawler type	Crawler type	Crawler type	Crawler type
Track width (mm)	450	500	450	500
Traction area (mm^2)	675000	1750000	675000	2060000
Load per unit area (kg/mm^2) in unload condition	0.00200	0.00204	0.00241	0.00155
5. Reaping				
5.1: Reaping mechanism	Reciprocating blade type	Reciprocating	Reciprocating	Reciprocating
5.2: Cutter bar				
Effective width (m)	1.5	2.0	1.5	2.2

Note: All studied models are tank type.

The actual average rate of harvester coverage, depending on the total time of operation, is known as the actual field capacity. The actual field capacity was calculated by dividing the area covered by the entire time according to the (Hunt,1995):

AFC= $\frac{A}{T}$

Where,

AFC= Actual field capacity, ha/h

A= Total covered area, ha

T= Total time of operation, h

Theoretical field capacity is the rate of field coverage of an implement that would be obtained if the machine were performing its function 100% of the time at the rated forward speed and always covered 100% of its width. It is also determined according to the (Hunt,1995):

TFC= $\frac{W \times S}{C}$

Where,

TFC= Theoretical field capacity, ha/hr

W= Cutting width of machine, m

S= Forward speed, km/h

C= Constant (Its value is 10)

Field efficiency is the ratio of effective field capacity and theoretical field capacity, expressed in percentages (Hunt,1995):

Ef= $\frac{AFC}{TFC} \times 100\%$

Where,

Ef= Field efficiency, %

AFC= Actual field capacity

TFC= Theoretical field capacity

Analysis

Data were analyzed as a single way factorial design (field length) according to Gomez and Gomez (Gomez and Gomez *et al.,*1984) using Statistix 10 programme (Statistix 10 software, 2013). Means were compared with the least significant difference (LSD) at which level of significant percentage test using Statistix 10 programme (Statistix 10 software, 2013).

RESULTS AND DISCUSSIONS

Field performance of the combine harvesters in Boro 2021-22 season

*Head feed combine harvester (*Kubota PRO588I-G*)*

The longer the field, the higher the forward speed, theoretical field capacity, practical field capacity, and field efficiency of the head feed combine harvester. Field efficiency for the head feed combine harvester was found to be 67.1 and 60.90% under the field length 61-70m in Sirajganj and Habiganj respectively, whereas field length ≤30m had the lowest field efficiency. Depending on the various field's length, forward speed and the effective field capacity in Sirajganj and Habiganj, which were varied significantly, ranged from 1.54 to 3.25 km/h and 1.3 to 3.56 km/h and 0.04 to 0.33 ha/h and 0.04 to 0.34 ha/h, respectively. Field efficiency did not vary significantly between the field lengths 51-60 and 61-70m in both the locations (Table 4). Traditional and medium-sized combine harvesters should operate at a forward speed between 3 and 6.5 km/h to work well when using a self-propelled combine (ASAE, 2009).

Table. 4. Field performance of the head feed combine (Kubota PRO588I-G**) harvester in the Boro, 2021-22 season.**

Field length type	Length, (m)	Total area (ha)	Forward speed, S (km/h)	Total operating time (h)	Effective field capacity (ha/h)	Theoretical field capacity (ha/h)	Field Efficiency (%)
Sirajganj							
L1	26	0.04	1.54	0.98	0.04	0.23	17.7
L2	39	0.10	1.82	0.98	0.10	0.27	37.4
L3	45	0.06	2.64	0.28	0.21	0.40	54.1
L4	52	0.09	2.93	0.33	0.27	0.44	62.1
L5	62	0.17	3.25	0.52	0.33	0.49	67.1
$LSD_{0.05}$	-	-	0.118	-	0.013	0.032	5.93
CV%	-	-	3.35	-	3.57	4.59	6.72
Habiganj							
L1	22	0.04	1.30	0.90	0.04	0.20	22.8
L2	32	0.07	1.61	0.85	0.08	0.24	34.1
L3	48	0.07	2.28	0.30	0.23	0.34	52.2
L4	56	0.07	2.98	0.35	0.20	0.45	58.5
L5	63	0.13	3.56	0.40	0.34	0.53	60.9
$LSD_{0.05}$	-	-	0.042	-	0.028	0.011	8.29
CV%	-	-	2.95	-	8.43	3.56	9.67

*Whole feed combine harvester (*Yanmar YH700*)*

The forward speed, theoretical field capacity, effective field capacity, and field efficiency of the whole feed combine harvester also increased significantly with the increase of the field length. In Sirajganj and Habiganj, field length 61-70m <30m determined to have the significantly higher field efficiency 62.6 and 68.1% for the whole feed combine harvester, whereas field length <30m had significantly lower field efficiency. The effective field capacity in Sirajganj and Habiganj varied significantly depending on the length of the different fields and was 0.25 to 0.87 ha/h and 0.13 to 0.87 ha/h, respectively (Table 5). Average harvesting speed and actual field capacity of the whole feed combine harvester (Zoomlion: 4LZT-4.0ZD) during Boro season in Haor region of Bangladesh were found 1.23 - 3.20 km/h and 0.15 ha/h (Islam, 2020). He also suggested to avoid the field sizes less than 800 m^2 for the Zoomlion combine harvester.

Table. 5. Field performance of the whole feed combine (Yanmar YH700**) harvester in the Boro 2021-22 season.**

Length type	Length (m)	Total area (ha)	Forward speed, S (km/h)	Total operating time (h)	Effective field capacity (ha/h)	Theoretical field capacity (ha/h)	Field Efficiency (%)
Sirajganj							
L1	28	0.02	4.32	0.08	0.25	0.86	28.9
L2	40	0.03	5.92	0.08	0.38	1.18	31.7
L3	50	0.08	6.43	0.12	0.67	1.29	51.8
L4	60	0.19	6.48	0.23	0.83	1.30	63.7
L5	65	0.20	6.95	0.23	0.87	1.39	62.6
$LSD_{0.05}$			0.1031		0.0794	0.1031	5.7447
% of cv			0.91		7.43	4.55	6.76
Habiganj							
L1	22	0.01	3.77	0.08	0.13	0.75	16.6
L2	38	0.03	5.20	0.08	0.38	1.04	36.1
L3	48	0.07	5.64	0.12	0.58	1.13	51.7
L4	55	0.17	5.71	0.23	0.74	1.14	64.7
L5	65	0.20	6.38	0.23	0.87	1.28	68.1
$LSD_{0.05}$			0.10		0.028	0.08	5.06
% of cv			1.03		2.76	4.16	5.64

Field performance of the combine harvesters in Aman 2022 season

*Head feed combine harvester (*Yanmar AG600A*)*

Field efficiency for the head feed combine harvester in Aman season was found to be 82.6 and 77.2% under the field length 61-70m in Sirajganj and Habiganj, respectively, whereas field length <30m had the significantly lowest field efficiency. Depending on the various field's length, the effective field capacity in Sirajganj and Habiganj, which were varied significantly, ranged from 0.192 to 0.52 ha/h and 0.19 to 0.51 ha/h while forward speed ranges from 2.98 to 4.3 km/h and 3.2 to 4.61 km/h, respectively (Table 6). Forward speed and effective field capacity of the head feed combine harvester (DR 150 A) were found 6.71 km/h and 0.33 ha/h, respectively in Amna season (Hasan *et al.,* 2019).

Table 6. Field performance of the head feed combine harvester (Yanmar AG600A**) in the Aman 2022 season.**

Length type	Length (m)	Total area (ha)	Forward speed (km/h)	Total operating time (h)	Effective field capacity (ha/h)	Effective cutting width, (m)	Theoretical field capacity (ha/h)	Field efficiency (%)
Sirajganj								
L1	24	0.05	2.979	0.250	0.19	1.5	0.45	44.8
L2	38	0.04	3.600	0.183	0.24	1.5	0.54	40.5
L3	42	0.09	3.844	0.217	0.39	1.5	0.58	71.9
L4	58	0.12	3.990	0.267	0.44	1.5	0.60	75.1
L5	65	0.08	4.307	0.150	0.52	1.5	0.65	82.6
$LSD_{0.05}$			0.1031		0.0827		0.0461	10.988
CV%			3.46		9.55		3.39	11.95
Habiganj								
L1	17	0.03	3.166	0.183	0.19	1.5	0.47	34.5
L2	32	0.06	3.388	0.267	0.24	1.5	0.51	44.2
L3	45	0.05	3.951	0.183	0.29	1.5	0.59	46.1
L4	55	0.11	4.097	0.267	0.42	1.5	0.61	67.0
L5	64	0.08	4.608	0.150	0.51	1.5	0.69	77.2
$LSD_{0.05}$			0.2455		0.0267		0.0322	7.6636
CV%			3.39		3.25		2.28	3.51

*Whole feed combine harvester (*FM World Ruilong*)*

Field efficiency for the whole feed combine harvester in Aman season was found to be 74.1 and 77.3% under the field length 61-70m in Sirajganj and Habiganj, respectively, whereas field length <30m had the significantly lowest field efficiency. Depending on the various field's length, the effective field capacity in Sirajganj and Habiganj, which are varied significantly, ranged from 0.32 to 0.90 ha/h and 0.29 to 0.70 ha/h respectively (Table 7). Forward speed, effective field capacity and field efficiency of the whole feed combine harvesters (new holland: CLAYSON 8080 and world star combine: WS7.0 PLUS) in large area were calculated 3.24 versus 4.10 km/h, 0.69 versus 0.53 ha/h and 64.3% versus 72.1% by (Suha Elsoragaby *et al.,* 2019.

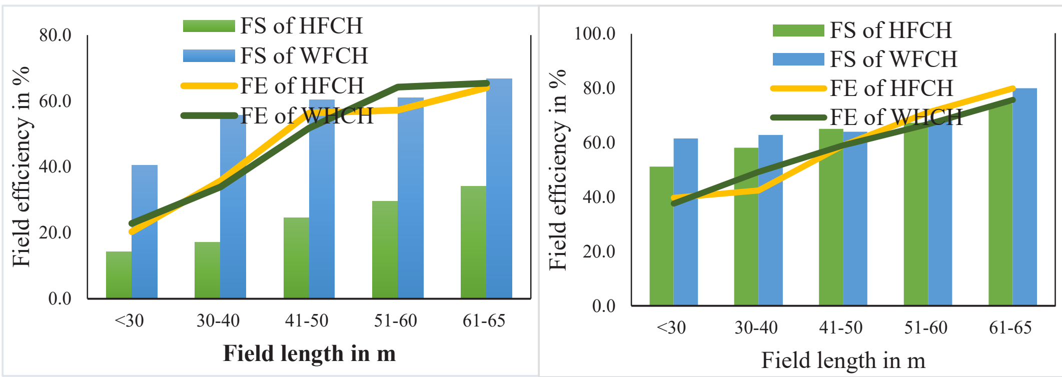
Table. 7. Field performance of the whole feed combine harvester (FM World Ruilong**) in the Aman, 2022 season.**

Length type	Length (m)	Total area (ha)	Forward speed (km/h)	Total operating time (h)	Effective field capacity (ha/h)	Effective cutting width (m)	Theoretical field capacity (ha/h)	Field Efficiency (%)
Sirajganj								
L1	29	0.058	3.773	0.183	0.32	2.2	0.83	38.2
L2	38	0.076	4.188	0.167	0.46	2.2	0.92	49.4
L3	44	0.088	4.659	0.15	0.59	2.2	1.02	57.2
L4	55	0.078	4.752	0.117	0.67	2.2	1.05	63.8
L5	62	0.075	5.54	0.083	0.90	2.2	1.22	74.1
$LSD_{0.05}$			0.1031		0.054		0.0639	5.1148
CV%			1.23		3.77		2.68	4.83
Habiganj								
L1	22	0.044	3.6	0.15	0.29	2.2	0.79	37.0
L2	39	0.078	3.343	0.217	0.36	2.2	0.74	48.9
L3	47	0.094	3.021	0.233	0.40	2.2	0.66	60.7
L4	54	0.08	3.471	0.15	0.53	2.2	0.76	69.8
L5	64	0.07	4.114	0.1	0.70	2.2	0.91	77.3
$LSD_{0.05}$			0.1203		0.0206		0.0734	2.3965
CV%			3.82		2.8		3.87	4.24

Forward speed and field efficiency of the head feed versus whole feed combine harvester

Forward speed and field efficiency increased with the increase of field length for the head feed and whole feed combine harvester in both Boro and Aman seasons while increasing rate varied with the machine type and seasons (Fig. 2). In Boro, 2021-22 season, forward speed of the head feed combine harvester increased 21 to 140% while it was increased 38 to 65% for whole feed combine harvester with the increase of field length based on the field length <30. Forward speed increased more for head feed combine harvester compared to whole feed combine harvester. In Aman, 2022 season, forward speed of the head feed combine harvester increased 14 to 45% while it was increased 2.2 to 30% for whole feed combine harvester with the increase of field length based on the field length <30m. Forward speed with the field length varied more for head feed combine harvester compared to the whole feed combine harvester because of complex mode of operation of the head feed combine harvester while overall variation was observed less in Aman season. Forward speed of the both type of harvesters was observed higher in Aman season as compared to Boro season which may be due to dryness of field during Aman season.

On contrary, field efficiency of the head feed combine harvester increased 76 to 215% and 7 to 101% during the Boro 2021-22 and Aman 2022 season, respectively with the increase of the field length based on field length <30m. It was increased 49 to 187% and 31 to 101% of the whole feed combine harvester during the Boro 2021-22 and Aman 2022 season respectively with the increase of the field length based on field length <30m. In Bangladesh, the average field efficiency of combine harvester is around 50% and varied from 30 to 60% with a field length of 30 to 65 m (Hossen, 2022). According to Phetmanyseng *et al.* (2019) harvesting efficiency of the combine harvester as affected by rice field size and other factors.



Boro 2021-22 season

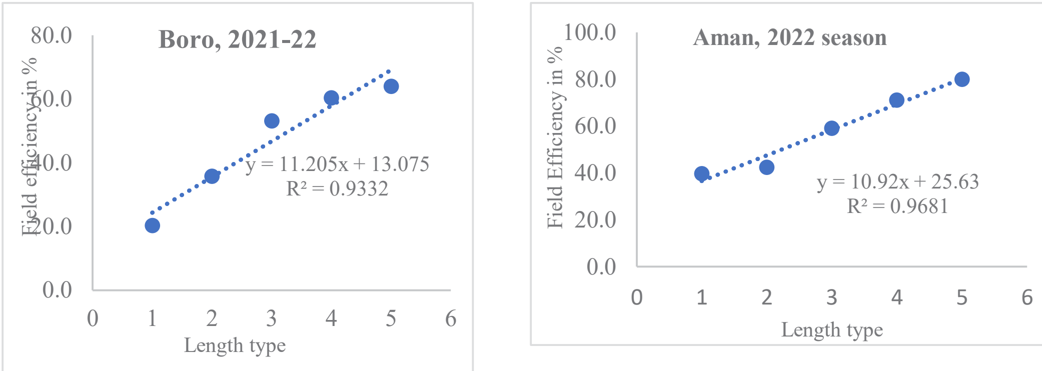
Aman 2022 season

Note: FS: Forward speed, HFCH: Head Feed Combine Harvester, WHCH: Whole Feed Combine Harvester, FE: Field Efficiency

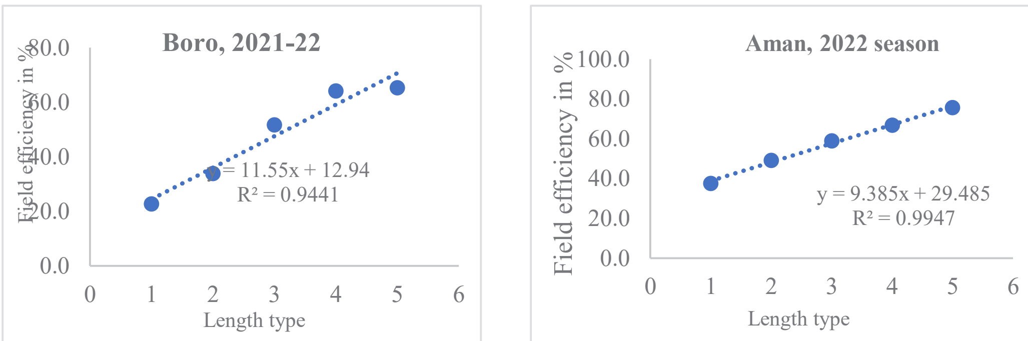
Fig. 2. Forward speed and field efficiency of the head feed versus whole feed combine harvester in Boro 2021-22 and Aman, 2022 season.

Effect of field length on field efficiency of the combine harvester

The field efficiency of both head feed and whole feed combine harvester was calculated with the length of the field which is presented in Tables 4-7. In all locations, the field efficiency of both the head feed and whole feed combine harvester increased with the increase in the field length. It is observed in the liner regression curve that the field performance of the head feed and whole feed combine harvester varied relatively in line with field length during Boro 2021-22 and Aman 2022 season respectively (R^2 = 0.93, R^2 = 0.94 and R^2 = 0.96 and R^2 = 0.99, respectively) (Fig. 3).



Head Feed Combine Harvester



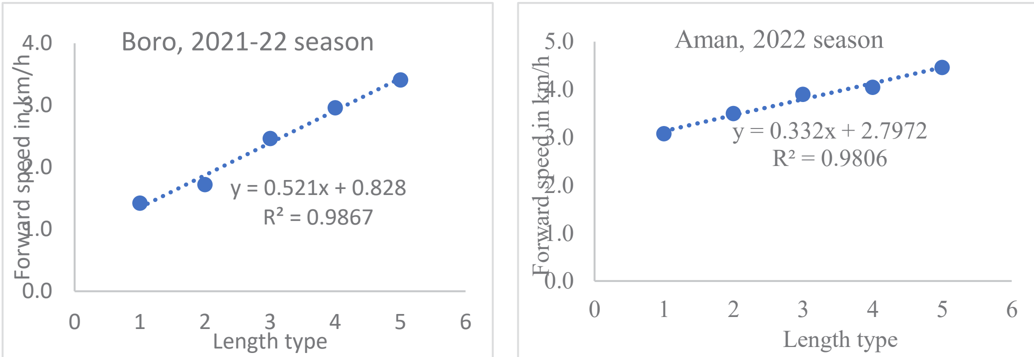
Whole Feed Combine Harvester

Note: Field length: L1≤30 m, L2: 31-40 m, L3: 41-50 m, L4: 51-60 m and L5: 61-70 m.

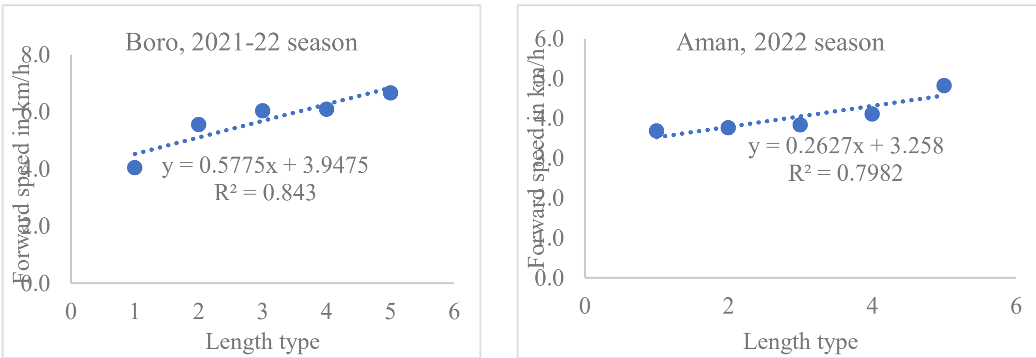
Fig. 3. Influences of field length on field efficiency of the combine harvesters.

Effect of field length on forward speed of the combine harvester

Forward speeds of both the types of combine harvester were determined with the length of the field which is presented in Table 4-7. It is observed in the liner regression curve that the forward speed of the head feed and whole feed combine harvesters varied relatively in line with field length in both Boro 2021-22 and Aman 2022 seasons respectively (R^2 = 0.91 and R^2 = 0.0.95, R^2 = 0.61 and R^2 = 0.95,) (Fig. 4).



Head Feed Combine Harvester



Whole Feed Combine Harvester

Fig. 4. Influences of field length on forward speed of the combine harvesters.

CONCLUSION

The study highlights the significant relationship between field size and the efficiency of combine harvesting operations. Larger field sizes tend to enhance operational efficiency by reducing the time and costs. Conversely, smaller fields lead to increased time and cost due to frequent turns and repositioning. Optimizing field sizes can lead to improved productivity and cost-effectiveness in harvesting practices using combine harvester, more specifically forward speed, effective field capacity, and field efficiency of both the head feed and whole feed combine harvesters varied linearly with the field size. The field length should not be less than 41-50 m for getting more than 50% field efficiency of the studied combine harvester in both the irrigated and non-irrigated seasons in Bangladesh. Additionally, recommendations for farmers and agricultural policymakers include considering field consolidation to maximize the benefits of mechanized harvesting. Further studies could explore the impact of other factors, such as crop type, logging condition, crop density, height of crop harvesting, field size and shape, plough pan depth, and on harvesting performance.

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AUTHOR’S CONTRIBUTION

M A Hossen generated the idea, design the experiment, developed the methodology, laid out the experiments, analyzed the data and prepared the report; Subrata Paul coordinated the field activities, S Islam and H Paul helped in data collection and tabulation for analysis according to the design; all authors read and approved the final manuscript.

AVAILABILITY OF DATA AND MATERIAL

Data used in this study are available from the first author upon request (dranwarhossenbrri@gmail.com).

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Effects of Climatic Variables on Aus Rice Production in Bangladesh Using Geo Statistical Techniques

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ABSTRACT

Bangladesh is a country with extreme weather conditions that is being investigated for its climatic effects on the production of rice (*Oryza sativa* L.) crops. Rice is cultivated as Aus, Aman, or Boro throughout the year in Bangladesh. The study was intended to explore the effects of climatic variables (rainfall, minimum temperature, and relative humidity) on Aus rice production in Bangladesh. Time series data of production, rainfall, minimum temperature, and relative humidity for the last decade (2011-2020) were collected from BBS (Bangladesh Bureau of Statistics), BRRI (Bangladesh Rice Research Institute), and BMD (Bangladesh Meteorological Department) to carry out the study. To make the research more specific and clear, the country was divided into four regions: the North- East, the North- West, the South-East, and the South-West. GIS (Geographic Information System) tools was used to prepare climatic mapping. The results show that rainfall and relative humidity had a significant impact on the production of Aus rice in the North-Eastern and North-Western regions, but rainfall was more effective than relative humidity in the North-Western part. Minimum temperature and relative humidity were the dominant variables in both the South-Eastern and the South-Western regions for Aus production.

Key words: Aus rice, Rainfall, Minimum Temperature, Relative humidity, GIS.

INTRODUCTION

Bangladesh is a developing country with an economic system based primarily on agriculture. The agricultural sector is significant to the country's overall economic growth and food security because of its high population density. The agricultural sector has historically played a significant role in Bangladesh (Molla *et al.* 2015). The country has an ideal climate and very fertile soil. So, a wide variety of distinct crops are produced here. The agriculture sector contributes about 13.02 percent in FY 2019–20 to the country`s Gross Domestic Product (GDP) and employs around 40.60 percent of the total labour force (BBS 2021). Rice (*Oryza sativa* L.) serves as their primary source of nutrition for more than 50% of the world's population. It also accounts for more than 60% and 25%, respectively, of Asia's and the world's cereal production and it accounts for over 30% of all the food consumed in Asia (Timmer, C.P. 2010). It is the primary source of nourishment for many of the world's highly populated nations, including China and Bangladesh, and is the main food item for a billion citizens of Asia (Jabran and Chauhan, 2015). According to Barua *et al.* (2014) it is the main food and a major proportion of the regular, balanced lifestyle of the Bangladeshi people. Rice is the staple food of around 167 million people in Bangladesh (BBS, 2017). In Bangladesh, the rice sector contributes half of the agricultural GDP and one-sixth of the national income (Elahi 2017). Rice is cultivated as Aus, Aman, or Boro throughout the year in Bangladesh. Aus, Aman, and Boro

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are successively grown from April to July, August to December, and January to May, respectively (Khondakar et al. 2022).

The climate is one of the most important factors affecting agricultural production. According to Ahmed and Alam (1999), variations in temperature and rainfall are becoming apparent due to climatic change both nationally and internationally. Floods, droughts, cyclones, and other dangers that are already more frequent in the country than ever before have been made worse by climate change and its variability. Flooding results from unpredictability in the timing and distribution of rain, while drought conditions are brought on by prolonged dry spells (Lai, et. al. 1998; Masud et al. 2014). According to projections, between 2005 and 2050, the average annual rice production in Bangladesh will decrease by 7.4% between 2005 and 2050, according to forecasts (Yu et al. 2010). Due to changing climatic situations, it has been predicted that reduction in rice productivity would become a significant issue in the future. (Yuji et al. 2009). Although technology has progressed, it has been suggested that the current patterns in climate element change may be too responsible for fully reversing yield trends (Islam et al. 2022). Since climate is the primary determinant of rice production, any changes in the climate have a significant impact on rice production (Huq et al. 1996; Karim et al. 1996; Yu et al. 2010). According to BRRI (1991), While Aman is nearly entirely rain-fed and grows in the monsoon months; Aus rice needs supplemental irrigation during the early stages of its growing season. Depending on the amount of rainfall available, Aman may also need supplemental irrigation during the flowering stage. In contrast, Boro rice is completely irrigated because it grows during the hot, dry winter and summer. (Mahmood, 1997). Only 5% of Aman rice and 8% of Aus rice are irrigated. (Ahmed, 2001; Sarker et al., 2019).

Research that aimed to investigate the relationship between climate variability (such as fluctuations in mean temperature, rainfall, relative humidity, and sunshine duration) and rice yields (such as Aus, Aman, and Boro rice varieties). The outcomes showed that humidity and rainfall have negatively affected Aus and Aman rice crops, while temperature and rainfall positively influence Boro rice yield (Islam et al. 2022). Another study was conducted to examine how Bangladesh's three different rice crops (Aus, Aman, and Boro) would be affected by climate change from 1972 to 2014. The results showed that for Aus rice, yield is positively correlated with rainfall and humidity but negatively correlated with maximum temperature. The statistically significant minimum temperature has no impact whatsoever on Aman production. The influence of the seasonal maximum average temperature is also seen to be identical and negatively connected to the yield of Boro rice (Chowdhury and Khan 2015). Different crops are affected by changes in climate variables in different ways. The average maximum temperature seems to make Aus and Aman rice production riskier while making Boro rice production less risky. The mean minimum temperature reduces the risk for Aus and Aman crops while increasing it for Boro rice. Last but not least, rainfall increases the risk of Aman rice while decreasing the risk of Aus and Boro rice (Sarker et al. 2014). The associations between the climate and crops were examined using national-level time series data. Findings showed that, except for Aus rice, the maximum temperature statistically significantly affected the production of all food crops. While the production and crop area of Aman rice were both considerably impacted by rainfall, the cropping area of Aus rice significantly benefited from it. According to statistics, humidity increased the production of Aus and Aman rice. Sunshine statistically significantly benefited only Boro rice yield (Amin et al. 2015). The production efficiency of Boro with environmental considerations is essentially identical to the production efficiency without environmental issues. The production efficiency of Aus and Aman is higher when environmental variables are used than when they are not. All types of rice production are significantly and favourably impacted by humidity. Given that temperature has a detrimental effect on production effectiveness, global warming may contribute to a decline in rice production efficiency. Only Boro production is positively impacted by rain (Hossain et al. 2013).

Aus rice varieties are typically short-duration, early-maturing varieties grown during the pre-monsoon season. In general, during April to July, which corresponds to the pre-monsoon and early monsoon period, temperatures in Bangladesh can range from warm to hot. Aus rice varieties are typically short-duration, early-maturing varieties grown during the pre-monsoon season. Minimum temperatures can range from around 20^{o}C to 28^{o}C. Minimum temperature can be important for maintaining favorable conditions during critical growth stages for Aus rice.

Besides, among the three crops, Aus, Aman, and Boro; Boro is the most significant and dominant crop, and total Boro production was 198,853 metric tons in FY 2020–21. The cultivation of Aman is also increasing day by day, and the total production for FY 2020–21 was 144,37,763 metric tons. But Aus's production is comparatively lower than that of Aman and Boro. The predicted total Aus production for the FY (2020–21) was 3,284.710 metric tons (BBS 2021). Farmers eventually started shifting to irrigated-Boro rice production, which was supported by its higher yields, and Aus rice started to lose its value. The production of rice in Boro is entirely dependent on irrigation, and the pressure of the groundwater is rising daily while the level of the water is falling. In contrast, Aus rice only needs 5% of the irrigation to be supplemented, and the groundwater pressure has to be lower than it is for Boro. Besides, the production of Aus rice has a significant relation with meteorological conditions such as temperature, precipitation, and humidity. It varies from region to region. For these reasons, it is necessary to have a sufficient understanding of Bangladesh's climate conditions and their changing patterns on a regional basis to boost Aus production.

A GIS (Geographic Information System) is a computer-based technology for storing, manipulating, analyzing, and retrieving data such as digital maps, images, or other data with spatial references, including latitude and longitude (BARC, 2001). With a small database, GIS and its accompanying maps and databases can serve a variety of general and specific functions. The ability of different data sets to be related via a shared spatial reference is an essential element of GIS (Zakaria et al., 2014). GIS (Geographic Information System) technology will help to visualize information on the maps. Because there has been no specific research conducted on a regional basis as well as using GIS for Aus.

The specific objectives of this study are: to identify the change in climatic conditions for Aus rice production using GIS mapping; to understand the impact of rainfall and minimum temperature and humidity on Aus rice (Aus) production; and to assess the change in Aus rice production under historical assumptions using trend analysis.

MATERIALS AND METHODS

Selection of study area

Secondary data on rainfall, temperature, humidity, and production were needed for the study's purpose. That's why the whole of Bangladesh was selected as the study area. Bangladesh is a South Asian nation with 64 districts that are located between 20°34' and 26°38'N and 88°01' and 92°41'E. Its total area is 147,570 km^2. The research would cover 64 districts in Bangladesh. 35 meteorological stations in Bangladesh were selected for the climate data. To make the research more specific and clear, the country was divided into four regions: the North- East, the North-West, the South- East, and the South-West.

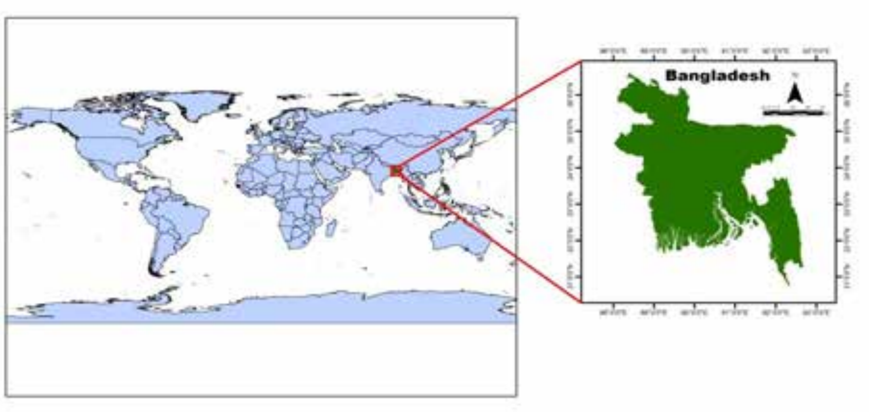

Fig. 1. Study Area: Bangladesh is selected as study area.

Districts under (Fig. 2)

- **North-Eastern Region:** Brahmanbaria, Dhaka, Gazipur, Hobiganj, Jamalpur, Kishoreganj, Manikganj, Moulavibazar, Munshiganj, Mymensingh, Narayanganj, Narsingdi, Netrakona, Sherpur, Srimangal, Sylhet and Tangail.
- **Sout-Eastern Region:** Bandarban, Chattagram, Cox's Bazar, Chandpur, Cumilla, Feni, Khagrachari, Laxmipur, Noakhali, Rangamati, Sitakunda
- **North-Western Region:** Rajshahi, Bogura, Rangpur, Dinajpur, Kurigram, Lalmomonirhat, Nilphamari, Panchagar, Thakurgaon, Gaibandha, Naogaon, Sirajganj, Chapai Nawabganj, Natore, Pabna, Kushtia, Meherpur, Jaypurhat
- **South-Western Region:** Faridpur, Madaripur, Chuadanga, Khulna, Jessore, Satkhira, Patuakhali, Barisal, Bhola, Jhenaidah, Magura, Narail, Gopalganj, Bagerhat, Barguna, Jhalakhati, Pirojpur, Rajbari, Shariatpur.

Thirty five meteorological stations of Bangladesh were selected for the climate data.

Stations under (Fig. 2)

- **North-Eastern Region:** Dhaka, Mymensingh, Sylhet, Srimangal
- **South-Eastern Region:** Chattagram, Cox's Bazar, Chandpur, Cumilla, Feni, Hatiya, Kutubdia, M.Court, Rangamati, Sandwip, Sitakunda, Teknaf
- **North-Western Region:** Tangail, Rajshahi, Bogura, Isaurdi, Rangpur, Dinajpur, Sydpur, Rangamati
- **South-Western Region:** Faridpur, Madaripur, Chuadanga, Khulna, Jessore, Satkhira, Patuakhali, Khepapura, Mongla, Barisal, Bhola

Fig. 2. Study Area: Regions and locations of meteorological stations.

Data Collection, Processing, and Tabulation Method

The information furnished in the study is based on a different database. The research was conducted using relevant secondary data from the Bangladesh Bureau of Statistics (BBS), Bangladesh Rice Research Institute (BRRI), and the Bangladesh Meteorological Department (BMD). The analysis was done for 10 years. Before 2011 there were less than thirty five meteorological stations in Bangladesh. Analyzing data prior to 2011 would not have any data coherency. To keep the analysis consistent, databases were made for the years 2011–2020. Data on Aus rice production (MT) from 2011-2020 was collected from BBS and BRRI, and climatic data on rainfall (mm), minimum temperature (^{o}C), and humidity (%) were collected from BMD for the following 10 years. To select the districts under the four regions, the map of Bangladesh was divided into four parts and manually named the North-Eastern, South-Eastern, North-Western, and South-Western regions.

The data entry (Aus production) for 64 districts was done using Microsoft Excel for 10 years. Data entry was also done for 35 meteorological stations for 10 years, and the database was updated on a daily basis. Microsoft Excel was used to calculate the average total annual rainfall, minimum temperature, and relative humidity. Furthermore, the Aus season runs from April to July. So, the following four months were selected from the meteorological database for rainfall, temperature, and humidity by filtering the whole database. After that, both production data and meteorological data were sorted into 4 regions (NE, SE, NW, and SW) using a pivot chart. A spatial analysis was done using ArcGIS 10.8 software for the meteorological dataset.

Using the spatial analyst tool in ArcGIS 10.8, maps of rainfall, minimum temperature, and humidity were created. After that, the last ten years of Aus rice production graphs were generated for each region, and they were then compared to statistics on rainfall, minimum temperature, and humidity. The effects of rainfall, minimum temperature, and humidity were then examined regionally for Aus rice production.

RESULTS AND DISCUSSION

Average Total Rainfall (Map and Trend)

In Bangladesh, average total rainfall maps (Fig. 3 and 4) for the ten years from 2011 to 2020 with a five-year gap have been prepared. The scale used to measure rainfall is in millimeters (mm). In different parts of Bangladesh, there have been numerous shifts in terms of the average annual rainfall. The average rainfall diagrams (Fig. 5) from 2011 to 2020 show Bangladesh's continuous rainfall variability. However, the regional graphs depict several scenarios of rainfall change.

Between 2011 and 2015 (Fig. 3), certain areas of Bangladesh, including Bandarban, Sylhet, Nilphamari, and Panchagar districts in the

South-East, North-East, and some parts of the North-West, experienced the highest rainfall exceeding 12000 mm. In the North-West, there was a 5000 mm decrease in average total rainfall, while Greater Barishal and Rangpur districts encountered moderate rainfall ranging from 7000 to 12000 mm

During 2016 to 2020 (Fig. 4), Sylhet in the North-East received the most rainfall exceeding 12000 mm. The North-West and South-West regions witnessed a 5000 mm decrease in overall average rainfall. Greater Dhaka, Cumilla, and Barishal districts saw moderate rainfall, ranging from 7000 to 11000 mm. These maps reveal a decline in the most rainy areas between 2016 and 2020 while witnessing an increase in regions with moderate rainfall.

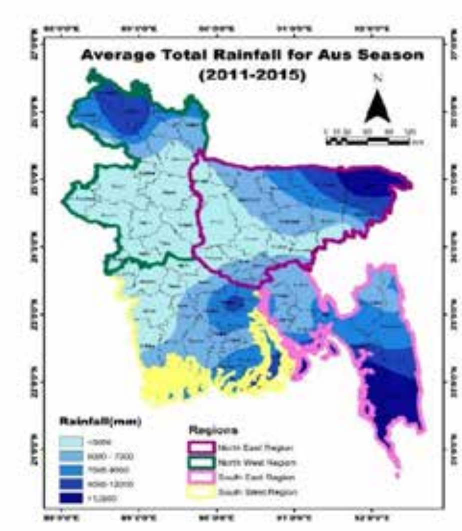

Fig. 3. Average Total Rainfall Map for Aus Season (2011-2015).

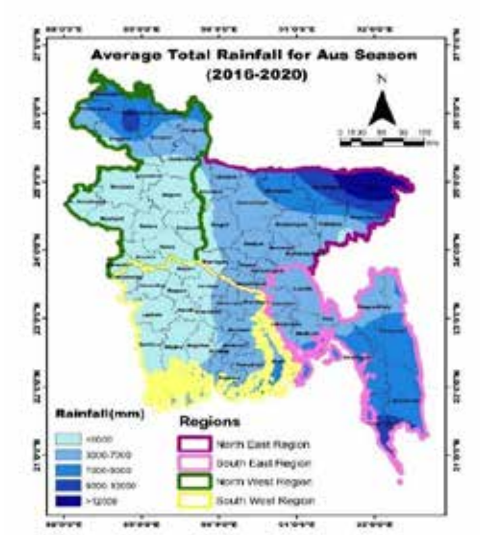

Fig. 4. Average Total Rainfall Map for Aus Season (2016-2020).

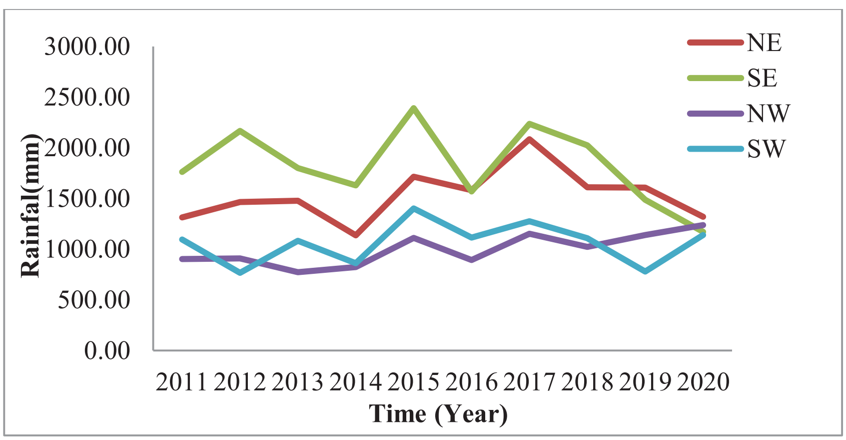

Fig. 5. Rainfall of the North-Eastern, South-Eastern, South-Western, and North-Western Region of Bangladesh

The graphs from 2011 to 2020 (Fig. 5) illustrate that Bangladesh's South-East region encountered the highest rainfall initially but experienced a decline after 2017. The trend line in the North-East region started as the second-highest but eventually rose to the top by 2020. The crossing of trend lines indicates fluctuation in rainfall amounts between 2011 and 2020, displaying periods of increase and decrease.

Average Minimum Temperature (Map and Trend)

Maps 6 and 7 show the average minimum temperature for the most recent decade, from 2011 to 2020, with a five-year gap. The scale of degrees Celsius (°C) is used to express temperature. The continuous temperature fluctuation in Bangladesh is depicted in the average minimum temperature curve (Fig. 8) from 2011 to 2020.

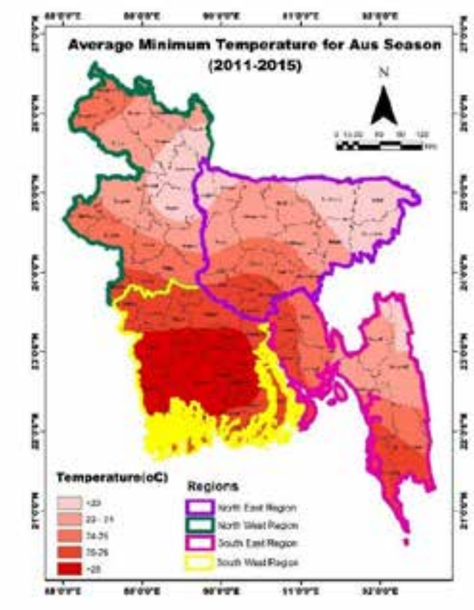

Fig. 6. Average minimum temperature map for Aus season (2011-2015).

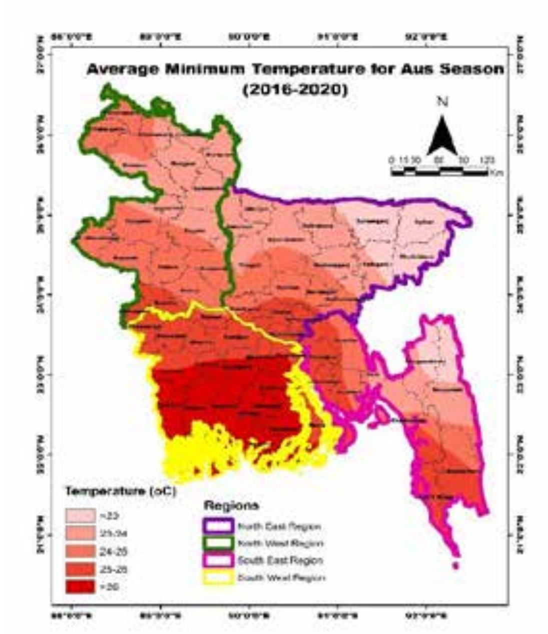

Fig. 7. Average minimum temperature map for Aus season (2016-2020).

Between 2011 and 2015 (Map 6), the South-West region of Bangladesh, encompassing greater Khulna and Barishal districts, recorded the highest average minimum temperature (>26°C). Meanwhile, the North-West and North-East regions maintained an average minimum temperature of 23 degrees Celsius.

During the 2016 to 2020 period (Map 7), a similar pattern persisted in Bangladesh's South-West region, with greater Khulna and Barishal districts experiencing the highest average minimum temperature (>26°C). Conversely, the North-West and North-East regions sustained an average minimum temperature of 23 degrees Celsius. Comparing these maps reveals that while the area with the maximum average minimum temperature remained relatively consistent from 2016 to 2020, there were notable changes in areas with moderate temperature levels.

The graphical representation from 2011 to 2020 (Fig. 8) highlights that the South-Western part of Bangladesh consistently displayed the maximum average minimum temperature, while the North-Eastern region experienced a decline in this measure. The trend lines indicate a rise in temperature for the South-Eastern region from 2012 to 2014, followed by a gradual decrease. Temperature patterns in the North-Eastern and North-Western regions fluctuated with recurrent increases and decreases, yet by the beginning of 2020, the average minimum temperature in the North-Eastern region began to decline.

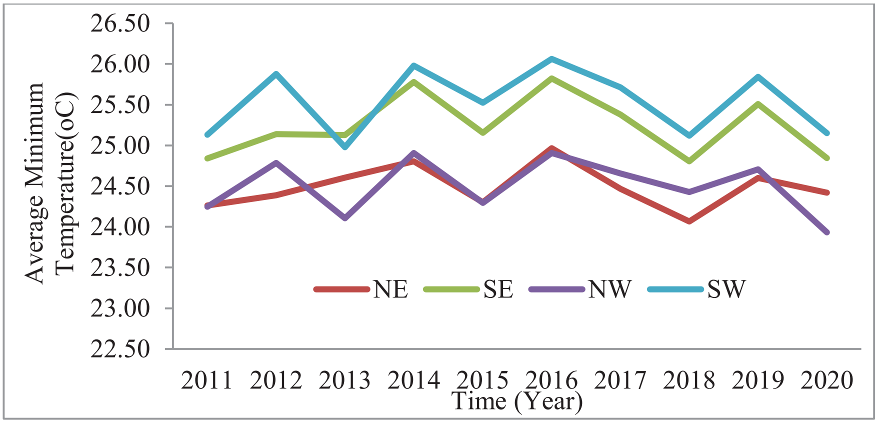

Fig. 8. Average minimum temperature of the North-Eastern, South-Eastern, South-Western, and North-Western region of Bangladesh.

Average Humidity (Map and Trend)

Average humidity maps (Figs. 9 and 10) have also been prepared for the last decade, from 2011–2020, with a five-year gap. The humidity unit is on a percentage (%) scale. The average humidity diagram (Fig. 11) from 2011–2020 shows the continuous fluctuation of humidity in Bangladesh.

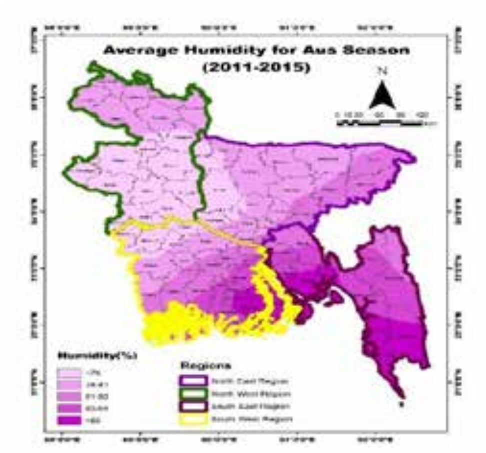

Fig. 9. Average humidity map for Aus season (2011-2015).

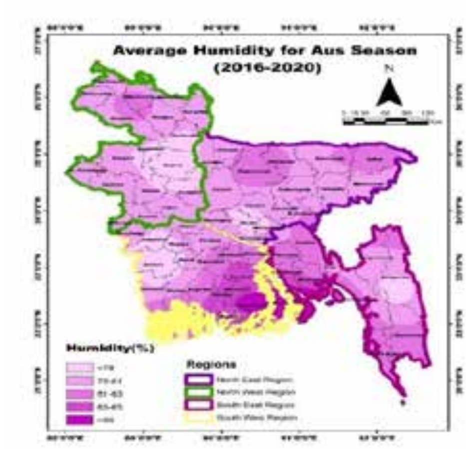

Fig. 10. Average humidity map for Aus season (2016-2020).

In the map from 2011 to 2015 (Fig. 9), the highest average humidity levels (>85%) were observed in the South-West and South-East regions of Bangladesh, encompassing Patuakhali, Noakhali, Bhola, Feni, and Bandarban districts. Conversely, the North-West regions experienced a decrease in average humidity, reaching 78%.

Similarly, according to the 2016 to 2020 map (Fig. 10), the areas with the highest average humidity (>85%) were situated in the south-western part of Bangladesh, specifically covering Patuakhali and Bhola districts. In certain districts across four regions, average humidity declined to 78%. A comparison between these maps reveals a change in the region with the maximum relative humidity between 2016 and 2020, along with an increase in the area experiencing minimum relative humidity.

The diagram representing 2011 to 2020 (Fig. 11) indicates that the South-East part of Bangladesh consistently maintained the highest average humidity, while the North-West region experienced decreased levels. The trend lines demonstrate that relative humidity predominantly prevailed in the South-Eastern region. The trend lines for the South-Western and North-Eastern regions overlap, converging to nearly the same level by the end of 2020.

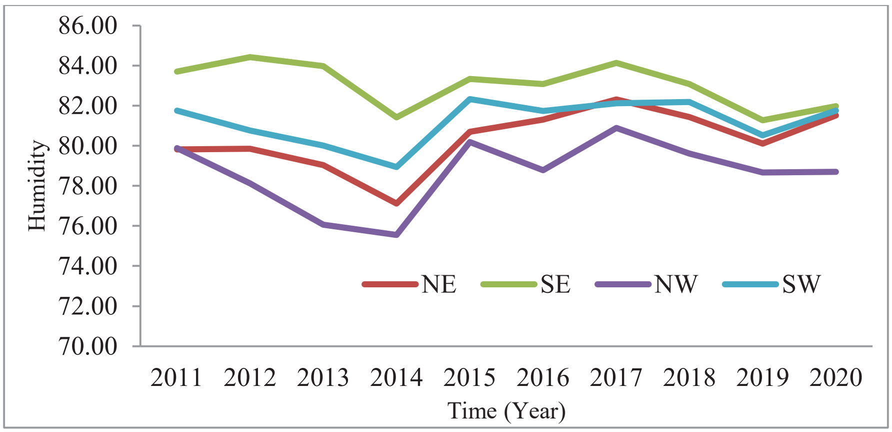

Fig. 11. Humidity of the North-Eastern, South-Eastern, South-Western, and North-Western region of Bangladesh.

Aus rice production in Bangladesh

In Bangladesh, different types of rice are grown depending on seasonal variations in the water supply. Since Aus, Aman, and Boro make up the majority of the nation's rice crop and grow in each of the three distinct seasons, rice is grown in Bangladesh all year round. The average planting and harvest dates for Aus are March or April and June or July, respectively (Sarker et al. 2012). The Aus rice production for each of the four dividing regions based on rainfall, temperature, and humidity has been estimated and shown. The production of rice in four regions of Aus is illustrated in figures from 12 to 15 For each graph, a linear trend line is constructed to illustrate the linear change in production. Figures 12 and 14 depict an upward trend in Aus rice output in the North-East and North-West regions over the past ten years, while a downward trend in production is seen in the South-East and South-West regions.

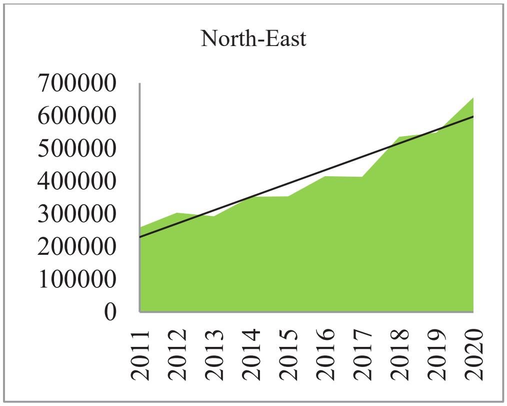

Fig. 12. Aus rice production and linear trend of the North-East region for the last decade.

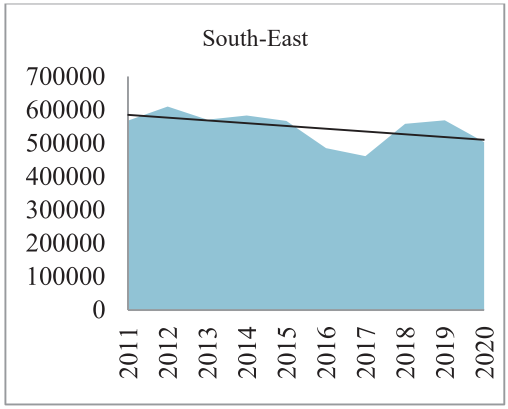

Fig. 13. Aus rice production and linear trend of the South-East region for the last decade.

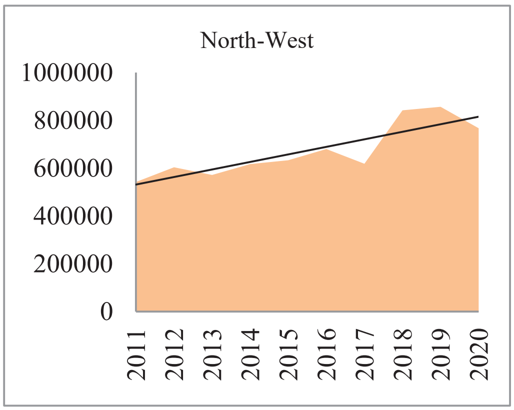

Fig. 14. Aus rice production and linear trend of the North-West region for the last decade.

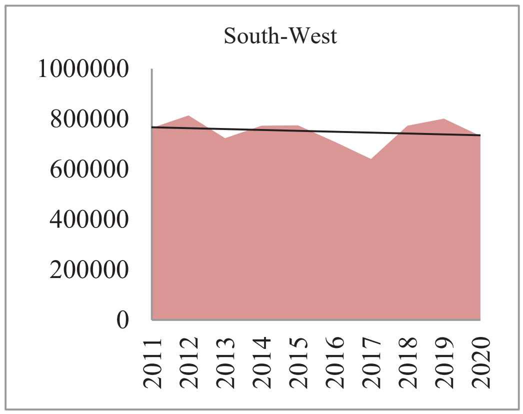

Fig.15. Aus rice production and linear trend of the South-West region for the last decade.

North-Eastern Region:

Brahmanbaria, Dhaka, Gazipur, Hobigonj, Jamalpur, Kishoreganj, Manikganj, Moulavibazar, Munshiganj, Mymensingh, Narsingdi, Netrakona, Sherpur, Srimangal, and Sylhet districts are included in the region.

The relationship between rice production and environmental factors like rainfall, Figures 16, 17 and 18 depicted temperature, and humidity in the region. The graph in Figure 16 illustrates that rice production has shown a consistent increase over time, correlating positively with higher rainfall levels. The lowest recorded rainfall occurred in 2014, followed by an upward trend from 2015 to 2017, a decline in 2018, and relatively stable levels from 2018 to 2019, before a decrease in 2020.

In contrast, the temperature graph (Fig. 17) displays an inverse trend concerning rice production, except for 2015 and 2017. The average minimum temperature initially increased from 2011, experienced a sudden drop in 2015, followed by fluctuations with a decrease in 2018 after rising in 2016 and 2017. Finally, temperatures rose again in

2019–2020. This fluctuating pattern in temperature doesn't significantly impact Aus rice production.

Fig. 18, showcasing humidity levels, exhibits an inverse trend between the periods of 2015–2017 and 2011–2013. Humidity trend lines were decreasing until 2014, then began to rise until 2019, slightly decreasing again in 2020. Despite these fluctuations, Aus rice production has increased over time. The analysis suggests that rainfall and humidity play dominant roles in influencing Aus rice production in this region. These variables exhibit a more consistent impact on production compared to the fluctuating pattern of temperatures, indicating a stronger correlation between higher rainfall/humidity and increased rice yields over time.

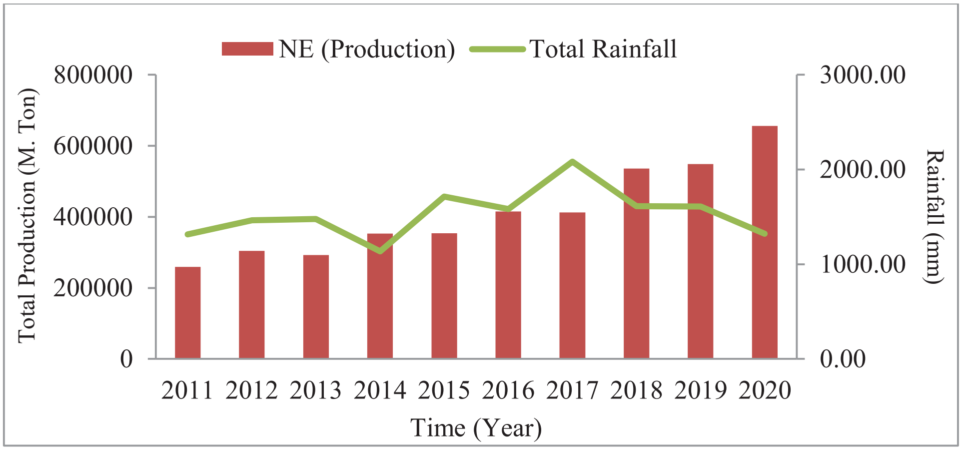

Fig.16. Combined graph of Aus rice production and rainfall in the North-Eastern region of Bangladesh for the last decade.

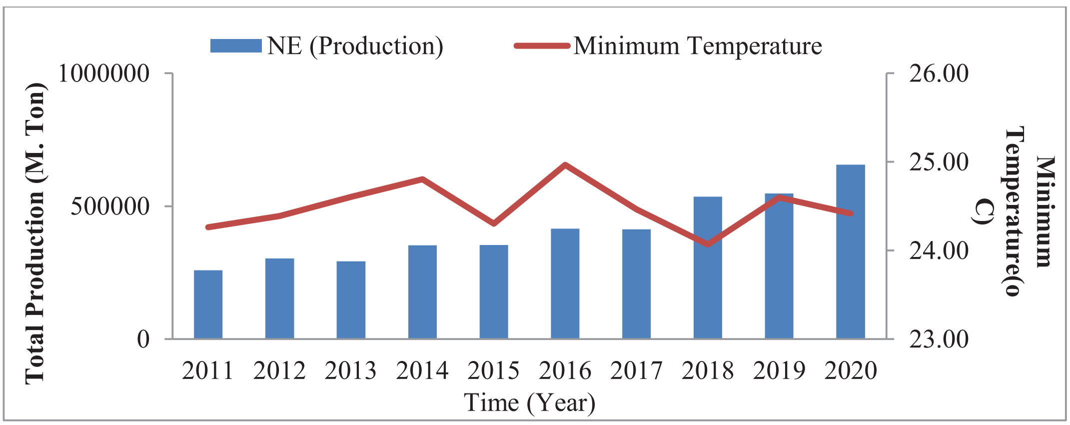

Fig. 17. Combined graph of Aus rice production and minimum temperature in the North-Eastern region of Bangladesh for the last decade.

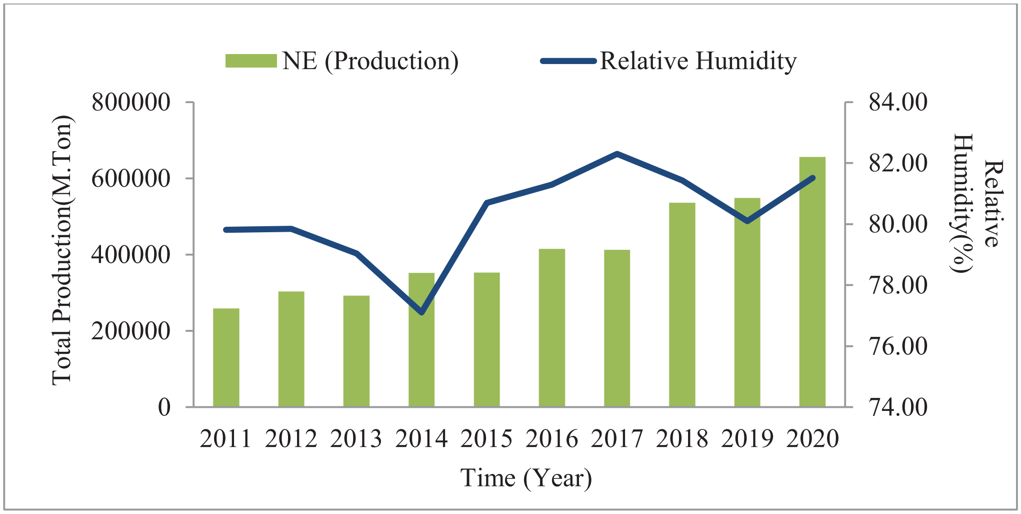

Fig. 18 Combined graph of Aus rice production and humidity in the North-Eastern region of Bangladesh for the last decade.

South-Eastern Region

Chattagram, Cox's Bazar, Chandpur, Cumilla, Feni, Khagrachari, Laxmipur, Noakhali, Rangamati, and Sitakunda districts are included in the region.

Figure 19 demonstrates a declining trend line until 2014, followed by an increase after 2016, and subsequently declining again from 2018 to 2020. Interestingly, there's little variation in production levels despite changes in rainfall, suggesting that rainfall has limited impact on the volume of Aus rice produced in this region. On the other hand, Figure 20, displaying temperature, indicates that production tends to increase with rising temperatures. However, production decreases when temperatures reach extremes, observed notably from 2016 through 2018 and 2020.

In Figure 21, the relative humidity graph shows an increase in both production and humidity until 2013. Despite a significant decline in humidity in 2014, production levels remained constant. Similar occurrences happened in 2017 and between 2017 and 2020. This graph, akin to the production vs. rainfall graph, suggests that humidity has a more pronounced impact on productivity compared to rainfall. Hence, average minimum temperature and humidity emerge as the dominant variables influencing Aus rice production in this region. Temperature fluctuations affect production, particularly when reaching extreme highs or lows, while humidity appears to have a more consistent influence on productivity.

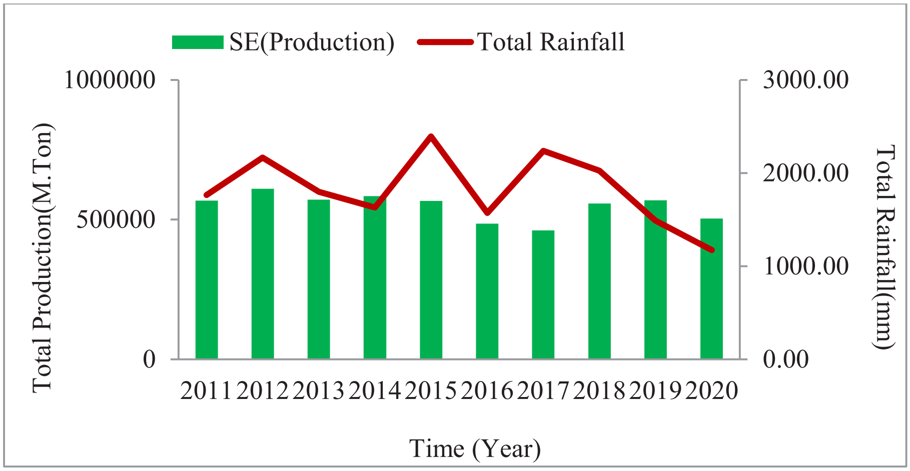

Fig. 19. Combined graph of Aus rice production and rainfall in the South-Eastern region of Bangladesh for the last decade.

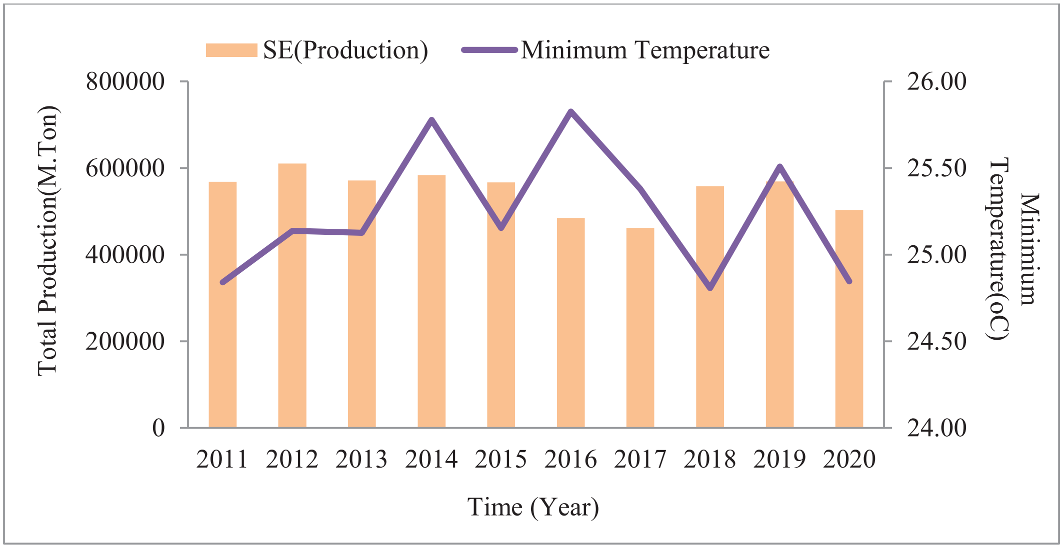

Fig. 20. Combined graph of Aus rice production and minimum temperature in the South-Eastern Region of Bangladesh for the last decade.

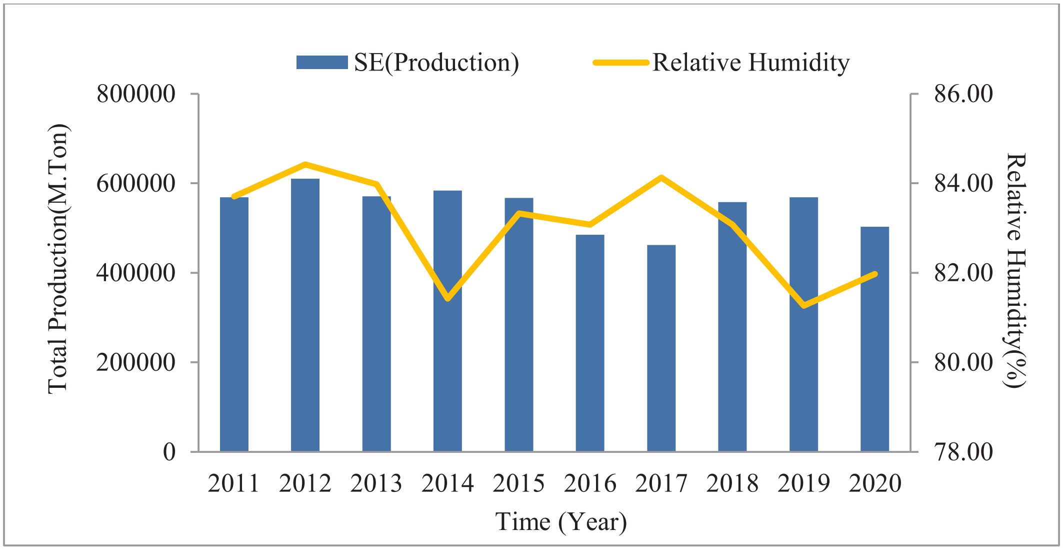

Fig. 21. Combined graph of Aus rice production and humidity in the South-Eastern Region of Bangladesh for the last decade.

North-Western Region

Tangail, Rajshahi, Bogura, Rangpur, Dinajpur, Rangamati, Kurigram, Lalmomonirhat, Nilphamari, Panchagar, Thakurgaon, Gaibandha, Naogaon, Sirajganj, Chapai Nawabganj, Natore, Pabna, Kushtia, Meherpur, and Jaypurhat districts are included in the region.

Figure 22 demonstrates an upward trend in rice production, attributed to increasing rainfall. Notably, Aus rice production peaks from 2019 to 2020, aligning with a higher trend line for rainfall during this period. The enhanced rainfall appears to positively impact Aus production, contributing to improved yields. Conversely, Figure 23, displaying temperature, doesn't display a discernible relationship between production trends in this area and changes in the average lowest temperature. Therefore, there seems to be no evident effect of temperature fluctuations on the production of Aus rice.

However, Figure 24, representing relative humidity, depicts a downward trend in humidity until 2014, followed by a subsequent rise. Concurrently, the level of production shows an upward trend. This observation indicates that relative humidity plays a significant role in Aus rice production in this region. Therefore, the analysis suggests that rainfall and humidity serve as the dominant variables influencing Aus rice production in this area. While increased rainfall positively impacts production, changes in humidity levels also have a substantial impact, with both factors contributing significantly to Aus rice yields. Conversely, temperature fluctuations do not seem to exert a notable influence on Aus rice production in this specific region.

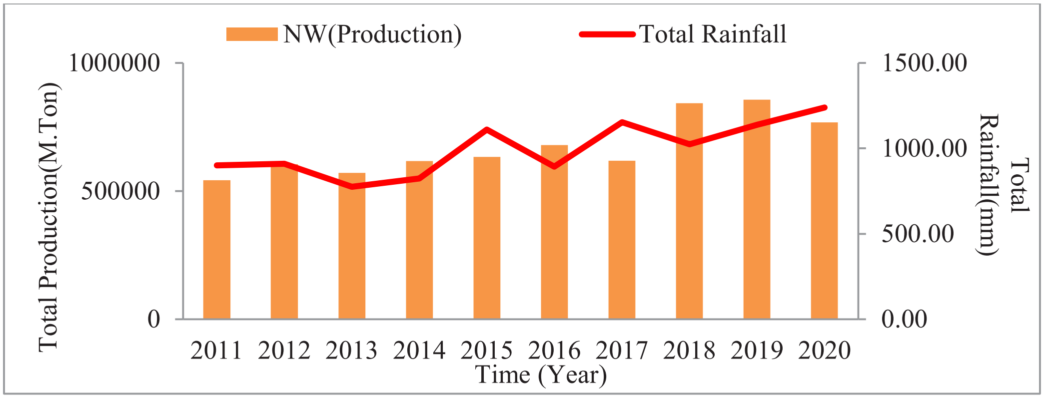

Fig. 22. Combined graph of Aus rice production and rainfall in the North-Western region of Bangladesh for the last decade.

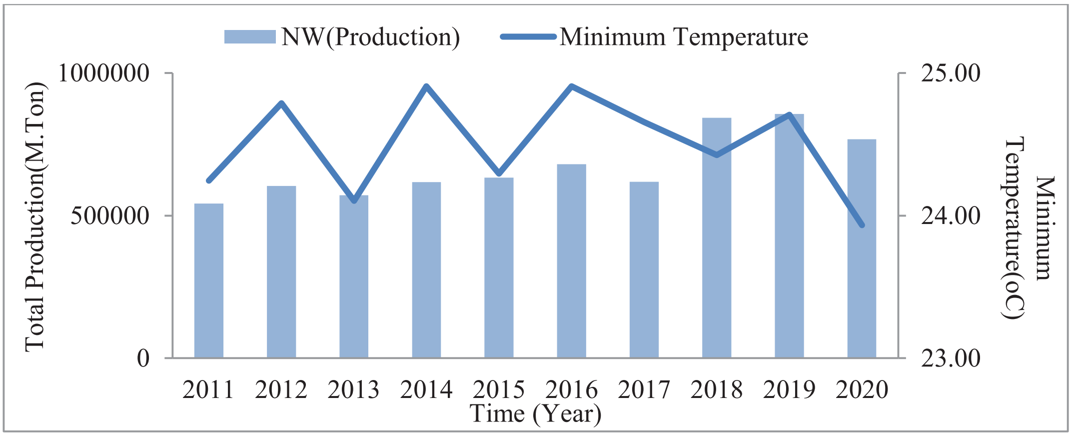

Fig. 23. Combined graph of Aus rice production and minimum temperature in the North-Western region of Bangladesh for the last decade.

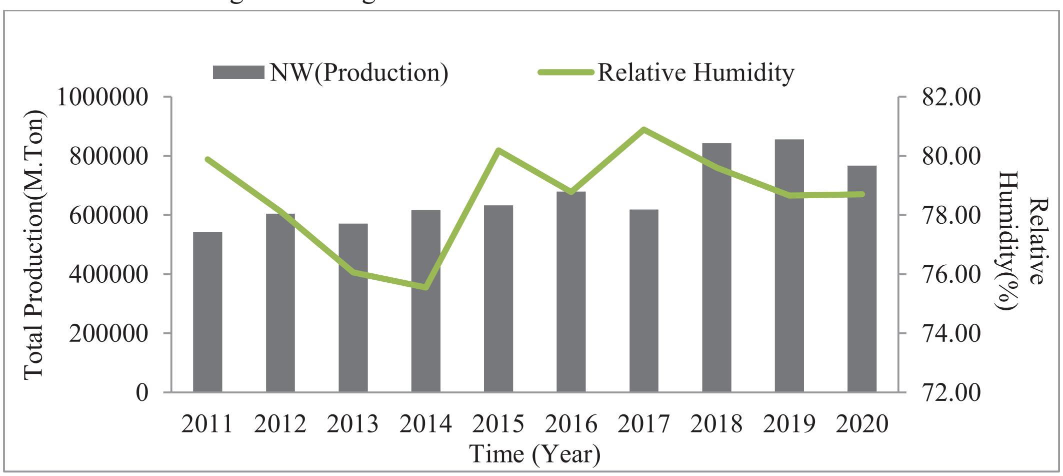

Fig. 24. Combined graph of Aus rice production and humidity in the North-Western region of Bangladesh for the last decade.

South-Western Region

Faridpur, Madaripur, Chuadanga, Khulna, Jessore, Satkhira, Patuakhali, Barisal, Bhola, Jhenaidah, Magura, Narail, Gopalganj, Bagerhat, Barguna, Jhalakhati, Pirojpur, Rajbari, and Shariatpur districts are included in the region.

Figure 25 illustrates a relatively stable Aus rice production trend over time despite fluctuations in the rainfall line, which mostly displays a downward trend. Surprisingly, these variations in rainfall seem to have limited effect on the production level of Aus rice in this context.

In Figure 26, the temperature graph reveals that Aus rice production is notably affected by temperature fluctuations in the years 2012, 2015–2017, and 2019. This suggests that, during these specific years, changes in temperature had an observable impact on the output of Aus rice.

Moreover, Figure 27, representing humidity, indicates a decline in humidity from 2011 followed by an increase after 2015. Until 2014, humidity did not seem to significantly impact the output of Aus rice. However, from 2015 through 2020, there was a noticeable influence of humidity on Aus rice production, implying that humidity levels began to play a more substantial role during this period. As a result, the analysis suggests that average minimum temperature and humidity emerge as the dominant variables influencing Aus rice production in this region. Temperature fluctuations seem to affect production in specific years, while humidity levels, particularly from 2015 to 2020, have a more pronounced impact on Aus rice yields. Despite fluctuations in rainfall, it appears to have limited effect on the production of Aus rice in this particular area compared to the significant influence of temperature and humidity on production levels.

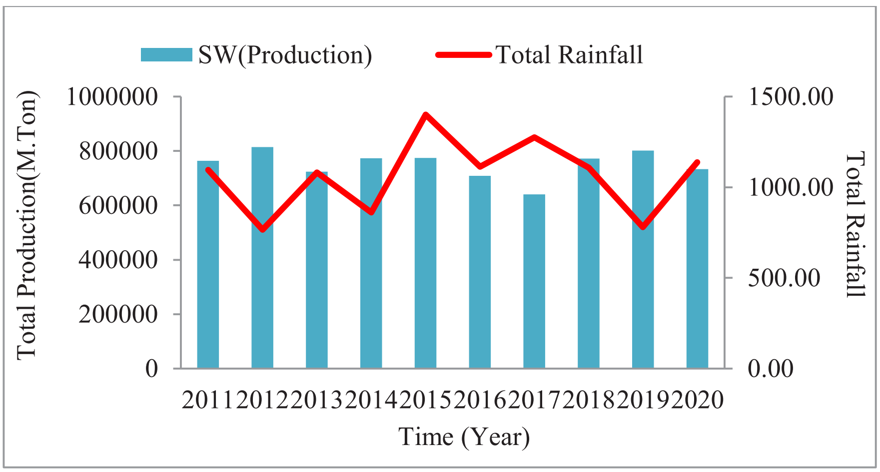

Fig. 25. Combined graph of Aus rice production and rainfall in the South-Western region of Bangladesh for the last decade.

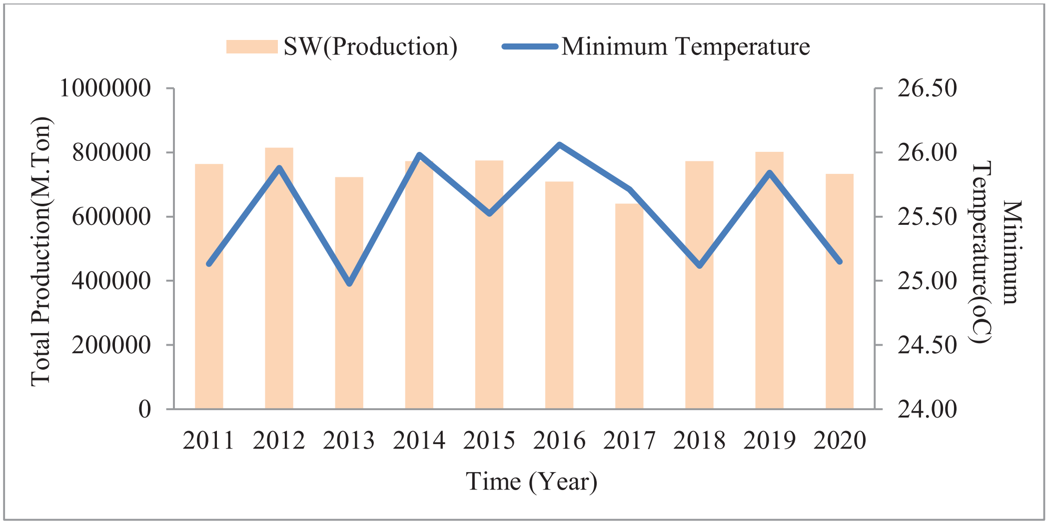

Fig. 26. Combined graph of Aus rice production and rainfall in the South-Western Region of Bangladesh for the last decade.

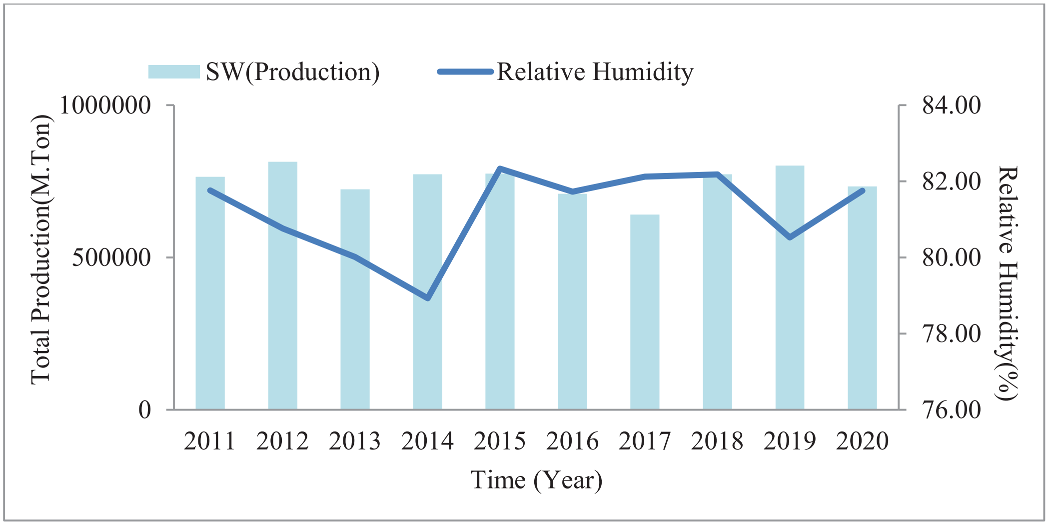

Fig. 27. Combined graph of Aus rice production and rainfall in the South-Western Region of Bangladesh for the last decade.

The impacts of climate change are evident in Bangladesh, with observable changes including rising temperatures, erratic rainfall patterns, and an escalation in extreme climate-related events such as floods, droughts, cyclones, sea-level rise, salinity, and soil erosion (*Asaduzzaman et al.*, 2010; *Yu et al*., 2010; Hossain and Deb, 2011). These factors notably affect the agricultural sector, especially rice production, occurring almost annually and often multiple times in a year (MoEF, 2005; *Yamin et al.,* 2005). Bangladesh's climate is characterized by high temperatures, heavy rainfall, elevated humidity levels, and distinct seasonal fluctuations (Sikder and Xiaoying, 2014).

Projections for the region indicate that climate change will contribute to increased circulation during the monsoon season, higher surface temperatures, and a heightened frequency and intensity of heavy rainfall events (Murshed et al., 2011). The Southwest agricultural region faces challenges during dry months due to water shortages and severe moisture stress (Faruque and Ali, 2005). Drought occurrences are frequent in Bangladesh's Northwest (Shahid, S. 2010). Moreover, there has been an observed increase in rainfall during the monsoon seasons in the western parts of the country (Shahid, S. 2010). Salinity intrusion poses a significant risk, particularly impacting the southern regions (Lal, M. 2011; Shahid et al., 2006). Crops heavily reliant on rainfall will be adversely affected by the year-to-year variation in rainfall, causing disruptions in irrigation systems (Shahid, S. 2011). These climate-related changes have substantial consequences on agriculture, particularly for rain-fed crops, posing challenges for food security and livelihoods in various regions of Bangladesh.

Region-wise Comparison of Aus Production

In addition, production in the Southern region was comparatively lower than in the Northern region. Eight districts were randomly selected from the area where production of Aus was high, and the comparison is shown in the following pie chart.

Table 1. Comparison of the Region-wise Aus Production.

Region	NE		SE		NW		SW	
District	Mymensingh	Sylhet	Chattagram	Noakhali	Rangpur	Rajshahi	Barishal	Khulna
Production (MT)	34338	197358	87523	80022	50325	134357	20507	6677
Percentage (%)	37.91		27.42		30.22		4.45	

Source: BBS, 2020

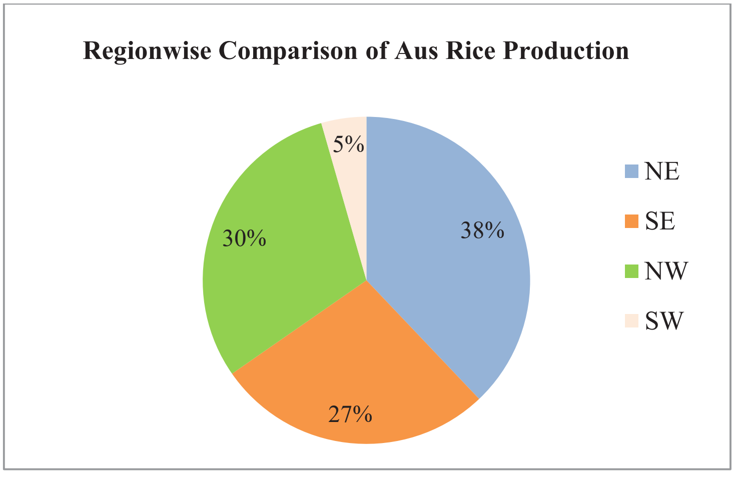

Fig. 28. Aus rice production region (Comparative analysis).

From the pie chart, in the North-Eastern region, production of Aus rice was 38% of the total selected area, and it was the highest among other regions. The North-Western, South-Eastern, and South-Western regions occupied 30%, 27%, and 5%, respectively.

The analysis of production vs. climatic variables (rainfall, minimum temperature, and humidity) graphs from the previous chapter reveals that rainfall and relative humidity had a significant impact on the production of Aus rice in the North-Eastern region. In the case of the South-Eastern part, minimum temperature and relative humidity were the dominant variables for Aus production. In the North-Western part, production of Aus rice followed the same pattern as the North-Eastern region, but the effect of rainfall was greater than the effect of relative humidity for this region, as rainfall followed an increasing trend from 2011 to 2020. Minimum temperature and relative humidity were important factors in the production of Aus rice in the south-western region.

CONCLUSION

The production of Aus rice in Bangladesh is significantly influenced by three key climatic variables: rainfall, minimum temperature, and humidity. However, their impact on Aus rice production varies across different regions in the country. Analysis of maps and trend lines indicates variations in these variables among regions, showcasing distinct influences on Aus rice production.

In the North-Eastern and North-Western regions, rainfall levels were relatively higher compared to the South-Eastern and South-Western regions. Conversely, the South-Eastern and South-Western parts experienced more favourable conditions in terms of minimum temperatures. Relative humidity was observed in all regions, particularly prominent in the South-Western part of Bangladesh.

Rainfall and relative humidity played a crucial role in augmenting Aus rice production in both the North-Eastern and North-Western regions. However, in the South-Eastern and South-Western regions, minimum temperature and relative humidity emerged as the primary factors influencing Aus rice production.

Interestingly, rainfall had a lesser impact on Aus rice production in the South-Eastern and South-Western regions compared to the other areas. Production levels were comparatively lower in the Southern region, which could potentially be attributed to the presence of salinity in the soil and water, along with the occurrence of natural calamities. Addressing salinity issues in soil and water could be crucial in increasing Aus rice production in these regions.

Moreover, the importance of minimum temperature seemed less pronounced in the North-Eastern and North-Western regions for Aus rice production. This could potentially be associated with the effects of climate change, drought, and CO_2 emissions. Further research is needed to comprehensively understand and address these challenges, aiming to enhance Aus rice production in these areas. Finding solutions to these issues through comprehensive research will be vital in sustaining and improving Aus rice production in Bangladesh.

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CONFLICT OF INTEREST

No potential conflict of interest was stated by the researchers (authors).

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