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Assessment of Bangladeshi Rice Landraces for Drought Tolerance Under Controlled Environment

M Khalequzzaman[1*] and N Haq[2]

ABSTRACT

Drought is one of the major challenging abiotic stresses for rice production in Bangladesh. This study investigated 48 rice landraces obtained from different Eco-zones of Bangladesh for their drought tolerance characteristics. Significant variation was observed in morpho-physiological traits such as length of root and shoot, dry weight of root and shoot, leaf stomatal resistance and stomata number. Stomatal conductance, root length, root dry weight showed significant correlation with visual drought score. A cluster analysis based on morpho-physiological characteristics identified six cluster groups and most of the promising drought tolerance landraces were found in cluster VI. Five landraces - Dhapa, Dud Kalam, Dular, Hogla Pata and Keora showed key desired characteristics for dry weight, stomatal conductance, evapotranspiration, water use efficiency and root parameters for drought tolerance. These landraces could be potential sources in breeding programme for the development of drought tolerant rice varieties.

Key words: Drought tolerance, landraces, morph-physiological traits, rice (*Oryza sativa* L.)

INTRODUCTION

Rice (*Oryza sativa* L.) is a major staple food for most of the people living in Asia and developing countries like Bangladesh. Due to its phylogenetic origin as a semi-aquatic plant (Das and Uchimiya, 2002), rice is highly dependent on a sufficient amount of water for its cultivation. As a result of increasing temperature and decreasing precipitation, drought has been one of the most significant constraints for crop productivity and, eventually, for global food security. Global warming and unpredictable rainfall patterns in recent years have led to excessive drought spells causing huge yield losses and excessive scarcity in food production in many parts of the world (Vikram *et al*., 2012). Amongst the abiotic factors that have created plant evolution, drought is considered as the most imperative and a major limitation for rice production in rainfed ecosystems (Nelson *et al*., 2014; Pandey and Shukla, 2015). Drought is a time span with low average precipitation/poor rain or higher evaporation rates causing a downfall in crop growth and yield (Rollins *et al*., 2013). About 20-25 million hectare of the world's potential cultivable rice land suffers from an inadequate supply of water and/or in drought conditions (Atlin *et al*., 2009). The severity and timing of water stress varied from season to season and year to year (Pantuwan *et al*., 2002). Its effect is often amalgamated with other abiotic and/or biotic stresses (Clover *et al*., 2001). It is estimated that about half of the world's rice is cultivated in rainfed and upland areas where unfavourable conditions limit crop production (Bennett, 2001; Pantuwan *et al*., 2002).

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The drought intensity and severity are very complex and it is dependent on different factors like frequency of rainfall, evaporation and soil moisture (Hao *et al.*, 2018; Oladosu *et al.*, 2019). In Asia alone, about 34 million ha of rainfed lowland and 8 million ha of upland rice exposed to drought stress (Singh *et al.*, 2016). Breeding for drought tolerance of rice plants have previously been attempted by a number of researchers, but progress has been slow because of complexity of the trait and lack of suitable donors with a high level of drought tolerance. Screening of thousands of genetic materials has been performed earlier for drought resistance in different corners of the world; however, only a few drought - tolerant varieties are yet recognized (Singh *et al.*, 2016; Kumar *et al.*, 2016). The main reasons for the minimal success were non-availability of truly drought-tolerant genotypes and lack of suitable screening methods (Pandey and Shukla, 2015).

The scientists from International Rice Research Institute (IRRI), Philippines have screened nearly 1000 rice accessions originated from 47 countries for drought tolerance and identified 65 accessions having drought-tolerance (Torres *et al.*, 2013). The identified tolerant rice varieties are either *aus* or *indica* type and the highest number of drought-tolerant accessions are *aus* ecotype (19) originated from Bangladesh, followed by India (7), whereas highest number of drought-tolerant *indica* accessions were originated from India (16) followed by Bangladesh (3) and Sri Lanka (3) (Torres *et al.*, 2013; Panda *et al.*, 2021). It is reported that the responses of rice plants to drought stress are believed to be complex that involves numerous physiological, biochemical and molecular changes (Upadhyaya and Panda, 2019; Gupta *et al.*, 2020; Melandri *et al.*, 2020).

The range of water stress is wide and can affect the crop at any stage of growth of rice plants.

As mentioned above that IRRI made some progress for the development of drought tolerant rice but the progress in developing drought tolerance rice varieties has been limited due to the influence of the environment and genetic interactions with them (Fukai and Cooper, 1995). Plant breeders have tried to interpret how environmental conditions influence genotypes, and how genotypes respond to different environments. Unfortunately, the influence of the environment over genotypes and yield models provides a little understanding of the biological significance of water stress (Turner *et al.*, 2001). It was reported that broader leaves play a role in better performance of indica rice under drought stress (Farooq *et al.*, 2010). Several leaf traits have been used for the screening of drought tolerant variety *i.e.* higher flag leaf area, leaf area index, leaf relative water content, leaf pigments content, stomata number etc. (Farooq *et al.*, 2009; Khalequzzaman, 2009, Mishra and Panda, 2017; Hussain *et al.*, 2018). Root characteristics of the plants are considered as the vital attributes for enhancing production under drought stress. Crop function under water stress is determined by the constitution and formation of rice root system (Panda *et al.*, 2021). In case of rice, the genotypes having profound root system, coarse roots, capacity of producing many branches and high root: shoot ratio is important characteristics for drought tolerance (Khalequzzaman, 2009; Kim *et al.*, 2020). It was reported that the morpho-physiological characteristics of rice roots play a major role in determining shoot growth and overall grain yield under drought stress (Kim *et al.*, 2020).

Several authors (Chakravorty *et al.*, 2013; Ray *et al.*, 2013, Hussain *et al.*, 2018; Panda *et al.*, 2021; Sabouri *et al.*, 2022) have investigated phenotypic diversity of rice landraces for adaptation to local conditions to different abiotic stresses. Different morpho-physiological parameters interact variously in relation to genotype in drought conditions. This paper aims to investigate rice landraces from different Eco-zones of Bangladesh with the goal of identifying drought tolerant rice accessions for rainfed and upland habitats. The obtained results may provide useful information for drought tolerance of Bangladeshi rice landraces for drought breeding programmes.

MATERIALS AND METHODS

A total of 48 rice accessions obtained from the Bangladesh Rice Research Institute (BRRI) genebank, which comprised of different agro-ecozones of Bangladesh, were used in the experiment. These accessions were chosen based on provenance information considered as the most suitable for drought tolerance experiments (Table 1).

Table 1. List of the rice landraces with accession number and provenance climates used in the study.

Accession	BRRI accession number	Growing season*	Collection district	Collection region***	Rainfall pattern[a]
Dharial	0018	Aus/Upland	DA-14**	-	Moderate rainfall
Dular	0022	Aus/Upland	DA-22	-	Moderate rainfall
HashiKalmi	0030	Aus/Upland	DA-23	-	Moderate rainfall
Kataktara	0039	Aus/Upland	DA-2	-	Moderate rainfall
Marichbati	0047	Aus/Upland	DA	-	Moderate rainfall
Panbira-1	0050	Aus/Upland	DA-12	-	Moderate rainfall
Hijolee	0571	Aus/Upland	Rangpur	Rangpur	Low/moderate rainfall
Aus Baku	1318	Aus/Upland	Kustia	Meherpur	Low/moderate rainfall
Hasha	1534	Aus/Upland	Dinajpur	JhikorGacha	Low/moderate rainfall
ManikMondal	1692	Aus/Upland	Faridpur	-	Moderate rainfall
HumaGambir	1738	Aus/Upland	Khulna	-	Moderate rainfall
Hanumanjata	1739	Aus/Upland	Khulna	-	Moderate rainfall
Boalia	2068	Aus/Upland	Kishoregonj	Hossainpur	Moderate/heavy rainfall
Ausa Bogi	2075	Aus/Upland	Kishoregonj	Kendua	Moderate/heavy rainfall
Agali	2082	Aus/Upland	Netrokona	Netrokona	Heavy rainfall
Bogi	2083	Aus/Upland	Netrokona	Netrokona	Heavy rainfall
Kumari Aus	2100	Aus/Upland	Jamalpur	Dewangonj	Heavy rainfall
Gopal Bhog	2109	Aus/Upland	Narsingdi	Narsingdi	Moderate rainfall
Sada Aus	2135	Aus/Upland	Pabna	Pabna	Low rainfall
Hazi Faram	2150	Aus/Upland	Rajshahi	Charghat	Low rainfall
Bina Muri-1	2181	Aus/Upland	Bogra	-	Moderate rainfall
Bina Muri-2	2184	Aus/Upland	Bogra	-	Moderate rainfall
Bakee	2358	Aus/Upland	Jamalpur	-	Moderate/heavy rainfall
Boila Bokri	3194	Aus/Upland	Munshigonj	Lauhajong	Moderate rainfall
Aus Nagra	3455	Aus/Upland	Jessore	Navaron	Low/Moderate rainfall
Aug Meghi	3456	Aus/Upland	Jessore	Navaron	Low/moderate rainfall
Bok Tulsi	3461	Aus/Upland	Shatkhira	Sadar	Moderate rainfall
Aus Kushi	3501	Aus/Upland	Shatkhira	Tala	Moderate rainfall
Bali Guri	3502	Aus/Upland	Mymensingh	Haluaghat	Heavy rainfall
Binna Toa	4197	Aus/Upland	Noakhali	Sonagazi	Heavy rainfall
Hogla Pata	3871	T. Aman	Barishal	-	Moderate rainfall
Kada Moni	0573	Aus/Upland	Rangpur	-	Low/moderate rainfall
Kala Mona	0984	T. Aman	Comilla	Baliaghata	Moderate rainfall
Kumra Gair	3878	T. Aman	Barishal	-	Moderate rainfall
Kacha Mota	3879	T. Aman	Barishal	-	Moderate rainfall
Kartik Sail	3662	T. Aman	Sherpur	Sherpur	Heavy rainfall
Kola Mocha	4141	B. Aman	Jhenidah	Jhenidah	Low/moderate rainfall
Lakhai	1800	Boro	Kishoregonj	Tarail	Moderate/heavy rainfall
Nuncha	0942	Aus/Upland	Khulna	Fakirhat	Moderate rainfall
Nona Balam	3203	T. Aman	Barishal	-	Moderate rainfall
Panbira-2	4150	T. Aman	Khulna	Fultola	Moderate rainfall
Tilock Kachari	0758	B. Aman	Chittagong	Boalkhali	Heavy rainfall
Aswina	0927	B. Aman	Sylhet	-	Heavy Rainfall
Dud Kalam	0278	T. Aman	Rangpur	Sundarganj	Low/moderate rainfall
Keora	0731	B. Aman	Comilla	-	Moderate rainfall
Hogla	4178	B/T. Aman	Jessore	-	Moderate rainfall
Kumari	0203	B//T. Aman	-	-	-
Dhapa	0320	T. Aman	Rangpur	Hatibandha	Low rainfall

[a]Low Rainfall <1600mm, Moderate Rainfall 1600-2500 mm, Heavy Rainfall >2500mm

- Data not available, * Rice growing ecosystem, Aus = Summer rice, T.Aman = autumn/rainfed lowland rice, Boro = winter/irrigated rice (Oka, 1991) **DA means Dhaka Agricultural station collection number ***Administrative unit

The accessions were screened in a glasshouse at the University of Southampton, England in 1999. The growing conditions were maintained with an average air temperature of 31°C, solar radiation of 18 MJ/m^2/day and relative humidity of 85% (Fig. 1).

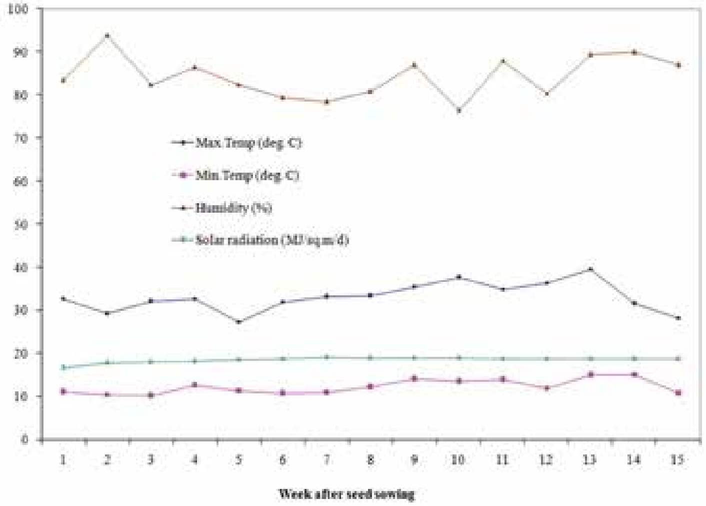

Fig. 1. The weather data in the glasshouse during the drought tolerance trial.

The experiment used a randomized complete block (RCB) design with four replications. Eight seeds of each accession were dibbled to 2 cm depth in pots. Each pot was 15 cm in diameter with a surface area of 177 cm^2, filled with 885 cm^3 of soil (John Innes compost number 2). The number of days required to seedling emergence was recorded. At seedling emergence, the pots were irrigated twice a week (200 cm^3/pot) until the seedling establishment. The seedlings were then thinned out to five seedlings per pot. After 45 days of seeding, irrigation was reduced to once a week until water stress was imposed. At the vegetative stage (before panicle primordial initiation), water stress was imposed after 45 days of emergence and visual score was made for the degree of drought intensity. The drought intensity was scored after 15 days of imposed stress using the methods described by O'Toole and Maguling (1981) and Standard Evaluation System of the International Rice Research Institute (IRRI, 1996). The drought severity scale was used from 0 to 9, where score of 0 signifies a score of higher drought tolerance (healthy leaves, no visible leaf rolling or necrosis) and score of 9 signifies high drought susceptibility (plants apparently dead). After measuring the visual drought score, pots were re-watered once a week.

The abaxial stomatal conductance (cm s^{-1}) of the youngest fully expanded leaf of five plants per treatment was measured using a diffusive resistance automatic porometer (Delta-T device, Mark II, Cambridge, England) just prior to the stress period (37 days after emergence) and at the stress period (52 days after emergence). The average stomata density (per mm^2) of five plants was measured using a stereomicroscope. Plant canopy heights were measured from the ground base to the end of the tallest leaf. Leaf area (cm^2) was calculated by using a Leica-Q-winimage processing and analysis system (Lecia-imaging system Ltd., Cambridge, England). After harvesting, plants were oven dried (constant weight at 80°C) and the weight of the shoots and roots of five hills per replication was measured. The ratios of root-to-shoot length (root/shoot length) and their dry weight (root/shoot dry weight) were calculated. Tiller numbers per hill from randomly selected five hills per replication were counted. Each pot was weighed once a week in order to determine water loss. Evapotranspiration (ET) was calculated weekly using a water balance equation: $ET = \{I + (S_1 - S_2)\}$/surface area of pot (mm/week); where I= irrigation, (S_1 and S_2) = the initial and final weight of pot, respectively. Water use efficiency (WUE) (g/kg) was then calculated as the ratio of dry matter or crop yield to ET (Kramer, 1980).

Analysis of variance and covariance, coefficient of variation (CV), simple linear regression and Pearson's bi-variate correlation were used to determine the inter-relationships between the drought-related parameters studied. A cluster analysis was then conducted using an agglomerative hierarchical clustering method (HCA), and a rescaled distance similarity measure was used to identify potential drought tolerant accessions. Principal component analysis (PCA) was carried out on the data matrix. The principal component scores with eigenvalues, which were greater than or equivalent to 1.0, were used as new variables for cluster analysis. All analyses were carried out using spreadsheets and SPSS version 10.

RESULTS

Table 2 presents descriptive statistics of the morpho-physiological traits of the accessions used in the experiment.

Table 2. Descriptive statistics and F values of different morpho-physiological characteristics based on two-way ANOVA in the tested 48 rice landraces.

Characteristic	Minimum	Maximum	Mean ±SE	CV (%)	F Value
Drought score	0.50	7.50	2.87±0.10	31.71	5.86**
Days to emergence	10.00	16.50	12.94±0.12	12.83	1.16^{NS}
Tiller number per hill	3.30	34.00	7.33±0.26	37.62	3.98**
Plant Canopy height (cm)	49.70	97.50	71.57±0.59	6.89	7.48**
Root length (cm)	15.50	40.44	25.81±0.66	14.62	4.11**
Root-shoot length ratio	0.21	0.63	0.36±0.01	15.88	4.94**
Stomatal conductance (cm/s) prior to stress	0.13	0.54	0.33±0.01	27.53	1.88**
Stomatal conductance (cm/s) at stress	0.05	0.11	0.08±0.00	80.50	1.05^{NS}
Stomata number	83	198	131±1.80	19.01	2.91**
Leaf length (cm)	32.50	60.10	45.09±0.38	7.30	6.18**
Leaf width (cm)	0.77	1.60	1.42±0.02	7.69	16.96**
Leaf area (cm^2)	13.47	38.50	22.53±0.48	12.58	9.37**
Root dry weight per hill (g)	0.40	2.80	1.22±0.03	21.16	4.42**
Shoot dry weight per hill (g)	3.20	33.90	6.54±0.11	17.76	3.31**
Total dry mass per hill (g)	3.60	36.30	7.76±0.11	15.66	3.09**
Root-shoot dry weight ratio	0.02	0.40	0.19±0.01	22.35	5.49**
Evapotranspiration (mm/day)	2.66	3.55	3.07±0.01	4.04	1.63*
Water use efficiency (g/kg)	3.41	12.54	6.93±0.10	14.48	2.88**

SE= Standard error of mean, CV= coefficient of variation ** Significant at the 1% level, *Significant at the 5% level, NS = Not significant at the 5% level, F value= Fisher significant test value.

Differences are observed in the parameters of mean, standard error and CV indicating the existence of diversity in the germplasm. Statistical distribution presented in Box and Whisker plot (Fig. 2) which showed the distribution of the population and dot sign in the middle of the boxplot indicate the mean performance of the traits.

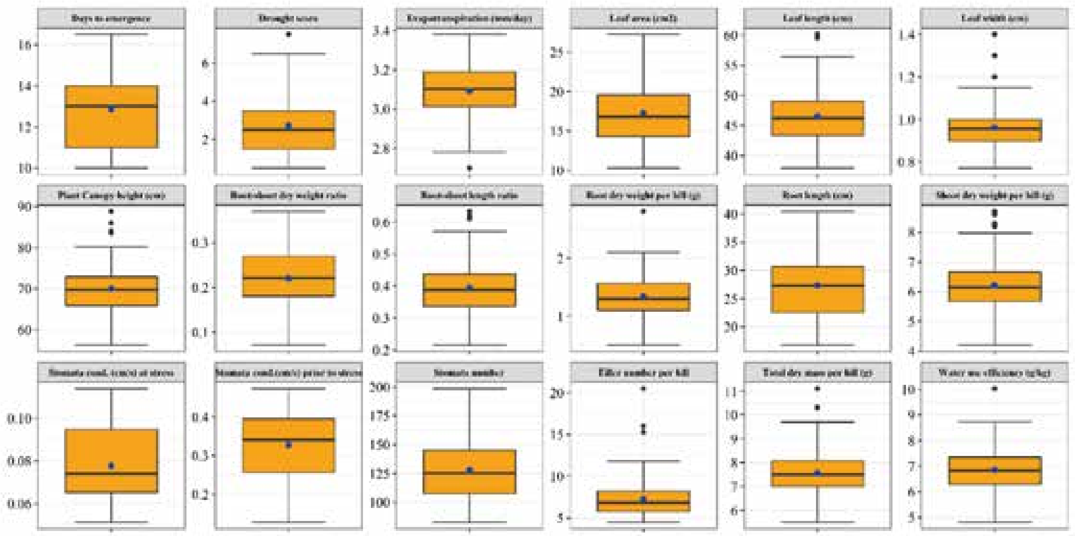

Fig. 2. Box and Whisker plot for different parameters of morpho-physiological traits of the tested 48 rice landraces.

Table 3 presents correlation between the studied parameters. Drought score showed significant negative correlation with root length (r = -0.43**), dry weight of root (r = -0.26**) and root-shoot length ratio (r = -0.33**). This indicates that larger root length and higher dry weight of roots are associated with drought tolerance of the accessions studied in this experiment.

A negative correlation also exists between drought score and stomata conductance prior to imposition of stress (r =- 0.27**) but it became significantly positive under stress conditions (r = 0.27**), whereas no relationships to stomata number. Thus, the accessions with desirable leaf characteristics, such as higher stomatal conductance when unstressed but lower conductance when stressed are potential drought tolerant traits.

It is apparent that the accessions with higher water use efficiency at lower evapotranspiration have useful characteristics for the selection of drought tolerant traits. The results revealed that the landraces Dhapa, Boalia, Dular, Hashi Kalmi, Hogla Pata etc had higher water use efficiency at lower ET (Fig. 3).

Table 3. Simple Pearson's correlation coefficient among the drought related parameters in the assessment of 48 rice landraces.

	Drought Score	St. cond. prior (cm/s) to stress	St. conduct (cm/s) at stress	Stomata number (per mm^2)	Leaf length (cm)	Leaf width (cm)	Leaf area (cm^2)	Shoot length (cm)	Root length (cm)	Root-shoot length ratio	Shoot dry weight (g)	Root dry weight (g)	Root-shoot dry weight ratio	Total drymass (g)	ET (mm/day)	WUE (g/kg)	Tiller number/ plant	Days to emergence
D. Score	1	-0.27**	0.27**	0.11^{NS}	-0.14^{NS}	-0.02^{NS}	-0.014^{NS}	-0.13^{NS}	-0.43**	-0.33**	0.06^{NS}	-0.26**	-0.28**	-0.01^{NS}	-0.14*	-0.01^{NS}	-0.09^{NS}	-0.25**
St. conductance prior to stress		1	-0.23**	0.32**	-0.02^{NS}	-0.001^{NS}	0.01^{NS}	-0.03^{NS}	0.25**	0.25**	-0.11^{NS}	0.29**	0.32**	-0.04^{NS}	0.24**	-0.02^{NS}	0.09^{NS}	0.04^{NS}
St. cond. At stress			1	0.11^{NS}	0.11^{NS}	0.09^{NS}	0.18*	0.07^{NS}	-0.26**	-0.26**	0.23**	-0.24**	-0.36**	0.16*	-0.002^{NS}	0.15^{NS}	0.02^{NS}	-0.03^{NS}
St. number				1	0.002^{NS}	0.06^{NS}	0.42**	0.02^{NS}	-0.25**	-0.23**	-0.02^{NS}	-0.04^{NS}	-0.02^{NS}	-0.03^{NS}	0.20**	-0.03^{NS}	0.04^{NS}	-0.10^{NS}
Leaf length					1	0.14^{NS}	0.36**	0.55**	0.07^{NS}	-0.21**	0.14^{NS}	0.04^{NS}	-0.07^{NS}	0.14^{NS}	0.05^{NS}	0.16*	-0.06^{NS}	0.07^{NS}
Leaf width						1	0.51**	0.48**	-0.14^{NS}	-0.36**	0.15*	-0.23**	-0.25**	0.08^{NS}	0.09^{NS}	0.08^{NS}	-0.12^{NS}	0.07^{NS}
Leaf area (cm^2)							1	0.45**	-0.17**	-0.38**	0.45**	-0.09^{NS}	-0.33**	0.41**	0.24**	0.41**	0.08^{NS}	0.01^{NS}
Shoot length (cm)								1	0.07^{NS}	-0.45**	0.23**	-0.02^{NS}	-0.14^{NS}	0.21**	0.17*	0.22**	-0.06^{NS}	0.13
Root length (cm)									1	0.85**	-0.07^{NS}	0.44**	0.47**	0.04^{NS}	0.02^{NS}	0.06^{NS}	0.14*	0.25**
R/Shoot length ratio										1	-0.15*	0.41**	0.47**	-0.05^{NS}	-0.04^{NS}	-0.04^{NS}	0.18*	0.18*
Shoot dry weight (g)											1	0.14*	-0.45**	0.97**	0.38**	0.94**	0.38**	0.26**
Root dry weight (g)												1	0.78**	0.37**	0.06^{NS}	0.39**	0.19*	0.10^{NS}
R/shoot dry weight ratio													1	-0.25**	-0.19**	-0.20**	-0.06^{NS}	-0.05^{NS}
Total dry weight (g)														1	0.37**	0.97**	0.41**	0.27**
ET (mm/day)															1	0.24**	0.23**	0.16*
WUE (g/kg)																1	0.38**	0.25**
Tiller number																	1	0.30**
Days to emergence																		1

D= Drought, St = Stomata, Cond= conductance, *Significant at $P<0.05$ and ** Significant at $P<0.01$, ^{NS}Not Significant at $P =0.05$ & 0.01, N=192.

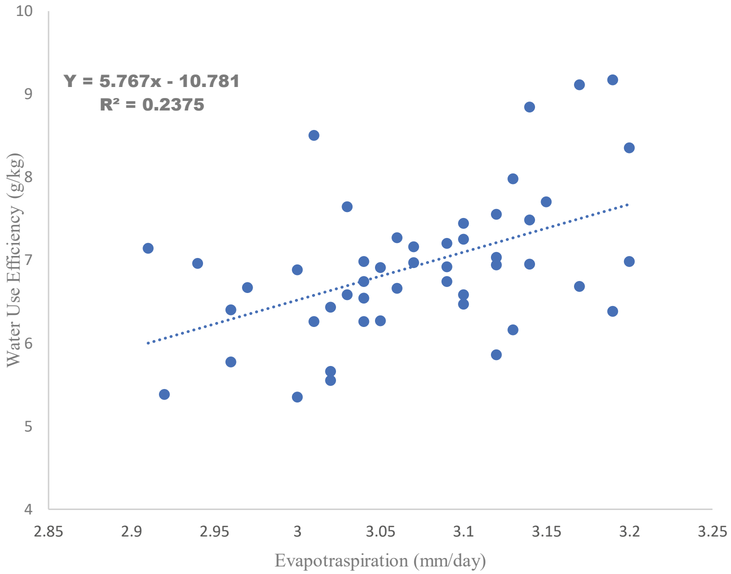

Fig. 3. Scatter diagram showing distribution of the tested 48 rice landraces based on evapotranspiration and water use efficiency.

Dry mass production was found to be related to ET and water use efficiency. The accessions- Dhapa, Dular, Boalia, Hashi Kalmi and Kala Mona had higher dry matter production with lower ET (Fig. 4, and Fig. 5). This is possibly due to their water use efficient characteristics. The above mechanism is considered useful for drought avoidance because the plants were able to reduce water loss by closing stomata.

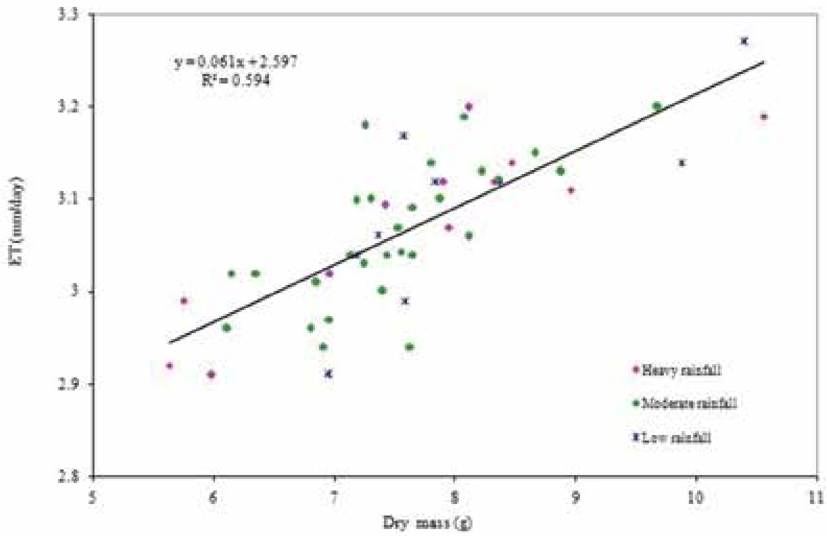

Fig. 4. Scatter diagram on the basis of ET (mm/day) and dry mass (g) showing distribution of the tested 48 rice landraces.

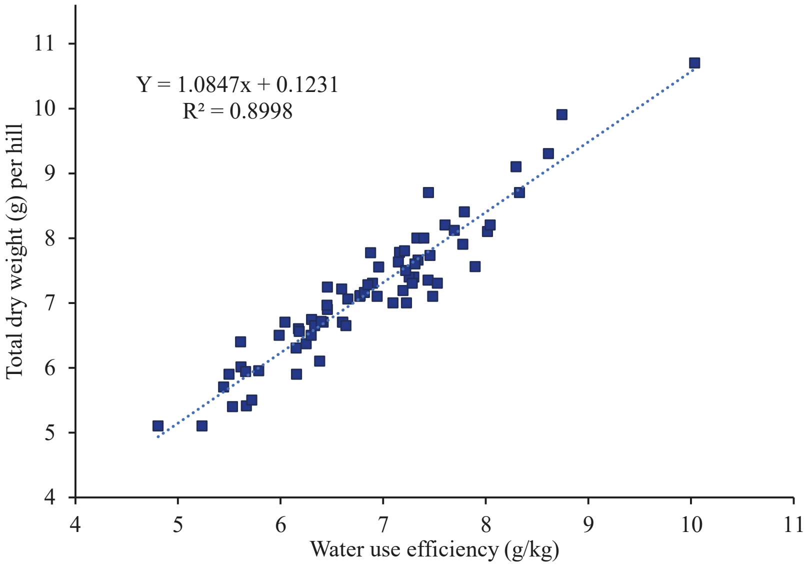

Fig. 5. Relationship between total dry weight (g) per hill and water use efficiency in the tested 48 rice landraces.

Dry matter results revealed that the accessions Dhapa (7.59 g), Kada Moni (7.18 g), Dular (7.63 g), Kala Mona (8.22 g), Dud Kalam (7.83 g), Keora (7.23 g) and Dud Mona (7.23 g) had increased dry matter production together with efficient water use characteristics (Table 4). Hence, these accessions could be selected for drought tolerance traits.

Table 4. Selected drought tolerant landraces with key characteristics related to drought tolerance.

Accessions	Drought score	Root length (cm)	Shoot length (cm)	Root dry mass (g) per pot	Shoot dry mass (g) per pot	ET (mm/day)	Total Dry weight (g)	WUE (g/kg)
Dud Kalam	1.56±0.7	33.58±2.8	63.32±2.6	7.52±1.0	31.62±1.9	3.12±0.1	7.83±0.3	6.94±0.2
Aug Meghi	1.63±0.6	30.87±3.3	78.43±1.5	7.13±1.1	30.50±2.9	3.07±0.1	7.52±0.5	6.97±0.5
Keora	1.75±0.5	29.97±1.9	65.89±3.6	7.28±1.0	28.89±4.1	3.03±0.1	7.23±0.7	6.58±0.7
Manik Mondol	1.75±0.5	30.25±1.9	74.55±3.2	6.16±0.5	28.38±2.5	3.05±0.1	6.91±0.5	6.27±0.5
Huma Gambir	1.75±1.0	30.42±1.7	76.93±9.3	6.74±0.5	31.00±3.5	3.04±0.1	7.55±0.7	6.98±1.0
Agali	1.75±0.5	29.08±4.4	79.95±6.4	6.20±0.3	30.91±5.2	3.09±0.2	7.42±1.0	6.74±0.7
Dhapa	1.75±0.2	34.12±2.3	68.76±2.5	8.06±0.6	29.90±1.5	2.91±0.2	7.59±0.4	7.14±0.1
Kala Mona	1.88±0.8	34.10±2.8	84.48±3.9	8.70±0.7	32.38±2.3	3.03±0.1	8.22±0.7	7.64±0.5
Bina Muri-2	1.75±0.5	27.78±4.9	60.90±3.1	5.44±0.8	25.25±1.2	3.02±0.1	6.14±0.3	5.67±0.2
Kada Moni	1.90±0.5	32.90±1.9	71.75±4.3	8.25±0.9	27.62±3.6	3.04±0.1	7.18±0.8	6.54±0.8
Bina Muri-1	2.00±06	26.55±5.1	61.25±5.4	5.64±1.0	24.88±1.9	2.96±0.1	6.11±0.5	5.77±0.3
Hogla Pata	2.00±0.4	27.55±2.6	69.20±2.5	6.00±0.4	28.00±2.6	2.96±0.2	6.80±0.5	6.40±0.3
Tilock Kachari	2.25±0.5	31.22±3.9	61.90±3.5	8.45±1.4	31.27±3.3	3.07±0.1	7.95±0.7	7.17±0.8
Dular	2.75±1.0	30.33±3.4	71.25±5.3	8.25±1.0	29.90±8.9	2.94±0.2	7.63±1.9	6.96±1.3
Hashi Kalmi	2.75±1.3	29.35±2.8	70.18±3.1	7.38±1.5	28.31±4.6	2.97±0.1	7.14±0.9	6.67±0.5

Principal component analysis (PCA) revealed six principal components, which indicates 85.1% of the variability of the parameters measured in this experiment Fig. 6 and Fig. 7 presents the bi-plot of the first two PCA's.

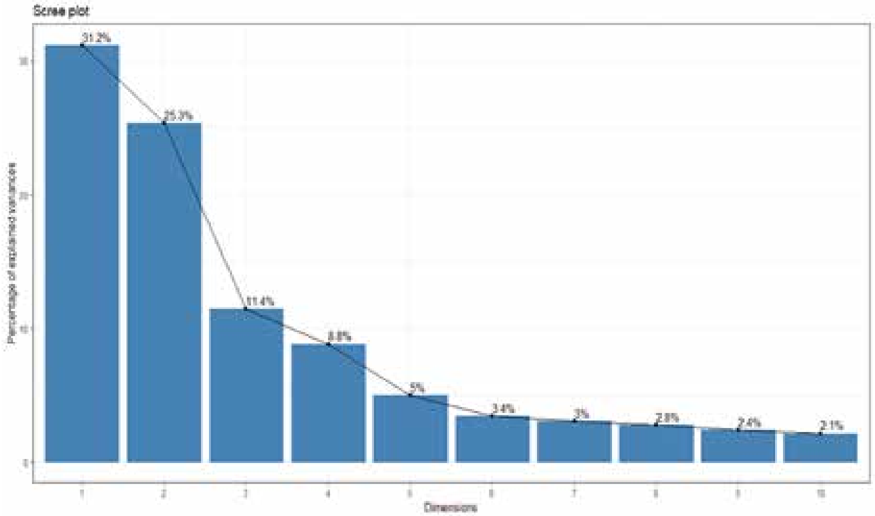

Fig. 6. Scree plot of PCA of the tested 48 rice landraces explained variance for different principal components.

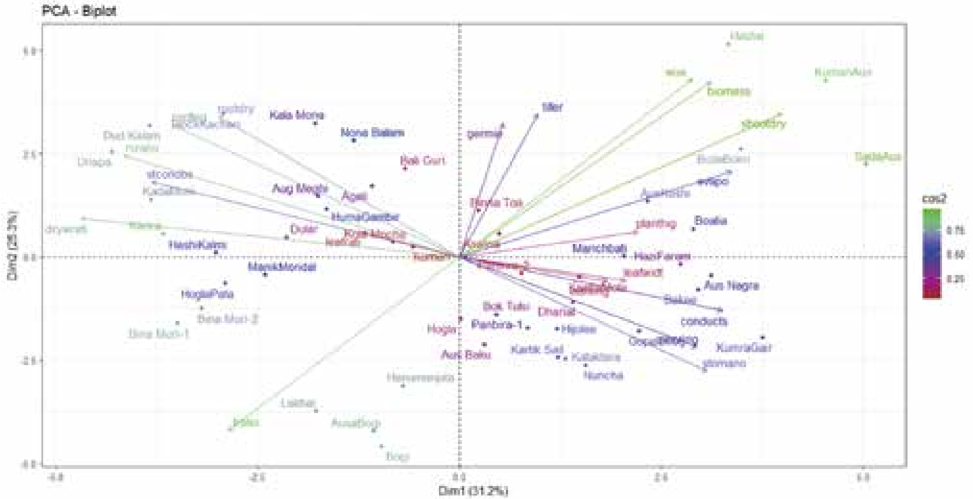

Fig.7. PCA of biplot for the tested 48 rice landraces based on morpho-physiological traits.

The hierarchical cluster analysis (HCA) was carried out to classify and identify accessions based on the variability extracted by principal components. The landraces with similar characteristics for drought tolerance were clustered into six groups (Fig. 8). Considering the studied results and findings, it is clear that the accessions with drought tolerance characteristics were grouped into cluster VI and identified drought tolerant landraces were Tilock Kachari, Dhapa, Hogla Pata, Keora, Dular, Hashi Kalmi and Dud Kalam (Table 5). In addition, this implies a probable genetic similarity among these accessions in terms of morpho-physiological characteristics and these are the potential candidate accessions for drought tolerance. Further evaluation is needed to confirm their drought tolerance.

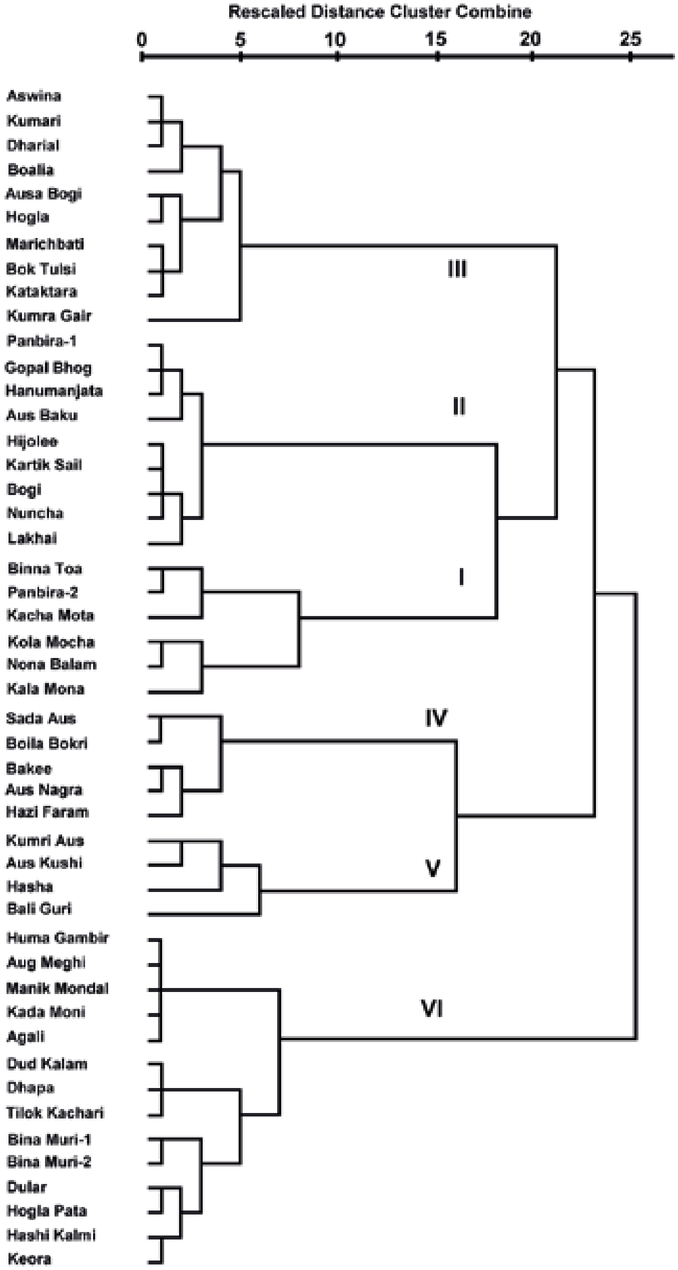

Fig. 8. Hierarchical clustering method (HCA) dendrogram showing different clusters of rice landraces based on morpho-physiological characteristics in the tested 48 rice landraces.

Table 5. Distribution of the tested 48 rice landraces revealed by cluster analyses based on morpho-physiological characteristics.

Cluster groups	Name of the landraces
I	Binna Toa. Panbira-2, Kacha Mota, Kola Mocha, Nona Balam, Kala mona
II	Panbira-1, Gopal Bhog, Hanumanjata, Aus Baku, Hijolee, Katrikshail, Bogi, Nuncha, Lakhai
III	Aswina, Kumari, Dharial, Boalia, AusaBogi, Hogla, Marichbati, Bok Tulsi. Kataktara, Kumar Gair
IV	Sada Aus, Bolia Bokri, Bakee, Aus Nagra, Hazi Faram
V	Kumari Aus, Aus Kushi, Hasha, Bali Guri
VI	Huma Gambir, Aug Meghi, Manik Mondal, Kada Moni, Agali, Dud Kalam, Dhapa, TilokKachari, Bina Muri-1, Bina Muri-2, Dular, Hogla Pata, Hashi Kalmi, Keora

DISCUSSION

As long as climate changes and subsequent substantial decline of grain yield caused by drought stress, the need for drought-tolerance is an essential breeding objective.

The visual drought score is one of the best selection indices for drought tolerance as this correlates with root development systems (Ingram *et al.*, 1990) and leaf water potential (O'Toole and Moya, 1978; Ekanayake *et al.*, 1985). In the present study, visual drought score showed significant correlation with dry root weight and stomatal conductance. The landraces/ accessions that were evaluated in this study showed a wide range of drought scores with physiological traits investigated. Correlations identified that deeper and more extensive root systems result in lower drought score (higher drought tolerance). This may be due to enhanced uptake of soil water. O'Toole (1982) states that rice cu1tivars with large root dry mass and deep roots can be considered to be drought tolerant. Similar results were reported by Chang *et al.*, (1974) in traditional upland rice cultivars. In the present investigation, it was found that the landraces, which had larger root systems and higher root-to-shoot ratios may be able to maintain high leaf water potential. The landraces which showed the above mentioned criteria may be selected for drought tolerance.

Stomata play an important role in regulating water loss. Drought score is related to leaf parameters such as stomatal conductance. The identified landraces might have the capability to maintain high leaf water potential and the ability to control transpiration through the opening and closing of the stomata. Further evidence from the leaf stomatal conductance confirmed this because the landraces/accessions with lower values of drought score had higher values of conductance prior to stress but considerably lower values under stress conditions. It has been reported by several authors (Collinson *et al.*, 1997; Clifford *et al.*, 2000. Azam-Ali and Squire, 2002, Khalequzzaman, 2002) that stomata decrease conductance under water stress conditions. This is in turn increases water use efficiency when water is limited (Clover *et al.* 2001). The present study show that stomatal conductance decreased to 28% under water stress conditions.

It is observed in the study that leaf rolling (one parameter used for drought score) decreases transpiration from leaves by reducing leaf area and along with stomata closure. This may have caused the genetical differences in maintaining high water status during water stress conditions. It has been reported that excessive transpiration is harmful for crop plants because it leads to significant reductions in productivity especially under limited water conditions

(O'Toole and Maguling, 1981). The crop must keep a careful balance between water uptake and loss to avoid excessive water deficit in the tissues otherwise it has to pay a yield penalty. This water balance may be achieved by a combination of reduced branching/tillering, leaf number, decreased leaf expansion and/or leaf rolling (Clifford *et al.*, 2000). In reality, leaf rolling helps to reduce the radiation incident on leaves and consequently reduces leaf temperature and water loss, which leads to increase the avoidance of dehydration.

In the current study evapotranspiration (ET) was related to the drought score and was significantly correlated with leaf area; root-shoot dry weight ratio, shoot and total biomass (Table 3). It appears that dry mass and water use efficiency increased with increased ET, although some landraces had higher dry mass production with low ET (Figs. 3, 4 and 5). This shows that these landraces have some capacity to increase their water use efficiency. This is possibly due to genetic factors as drought has minor effect on water use efficiency (Clover *et al.*, 2001).

It was reported earlier that longer roots, greater root dry weight and higher ratios of root-shoot length to dry weight can play a significant role in drought resistance mechanisms (Ludlow and Muchow, 1990; Thanh *et al.*, 1999; Azam-Ali and Squire, 2002; Khalequzzaman, 2002; Khalequzzaman, 2009). Several landraces of this studied showed that root-to-shoot ratios and leaf stomatal conductance prior to stress conditions reduced under stress conditions, indicating tolerance to drought. Principal component analysis (Figs. 6 and 7) and cluster analysis revealed that most of the possible drought tolerant accessions were confined to one group (Table 5 and Fig. 8) This implies that they were similar in relation to their morpho-physiological characteristics such as root system, root biomass, dry weight of root and shoot, stomata number and conductance.

To solve the water scarcity issue, developing drought-tolerant rice varieties has been a major challenge of rice breeders. Moreover, utilizing best-performing genotypes as donor parents in breeding program could be very effective in improving rice for drought tolerance.

CONCLUSION

In conclusion, the assessment of Bangladeshi rice landraces for drought tolerance under controlled environment has provided valuable insights into the potential of traditional varieties to withstand drought stress. The results of this study suggest that some of these landraces have adaptive traits that enable them to cope with limited water availability, which is a promising indication for their potential use in future breeding programs aimed at improving drought tolerance in rice cultivars. The evaluation of physiological and morphological traits provided a comprehensive understanding of the response of these landraces to drought stress. It was observed that some of the landraces maintained their growth under drought conditions, indicating the existence of natural variability that can be exploited to improve rice cultivars' resilience to water scarcity. The findings of this of this study have significant implications for sustainable agriculture in Bangladesh, where drought stress is a prevalent constraint for rice production. The use of drought-tolerant varieties can help farmers cope with changing climatic conditions and reduce their reliance on irrigation water, ultimately contributing to food security and rural livelihoods. Overall, the assessment of Bangladeshi rice landraces for drought tolerance under controlled environment provides a promising direction for future breeding programmes and highlights the importance of preserving traditional crop diversity as a valuable resource for agricultural sustainability.

AUTHORS' CONTRIBUTION

NH generated the idea and MK developed methodology, gathered data, carried out analysis and wrote the manuscript.

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DECLARATION OF INTERESTS

We wish to confirm that there are no known conflicts of interest associated with this publication.

REFERENCES

Atlin, G, N R Venuprasad, J Bernier, D Zhao, P Virk, and A Kumar. 2009. Rice germplasm development for drought-prone environments: progress made in breeding and genetic analysis at the International Rice Research Institute. *In*: Drought frontiers in rice: crop improvement for increased rainfed production, R Serraj, J Bennett and B Hardy (Eds.). Singapore: *World Scientific publishing and International Rice Research Institute*, Los Banos, Philippines, 35-60.

Azam-Ali, S N, and G R Squire. 2002. Principles of tropical agronomy. *CAB International*, UK and USA.

Bennett, J. 2001. Summing up: cutting edge science for rice improvement breakthroughs and beneficiaries. *In*: Rice Biotechnology: Improving yield, stress tolerance and grain quality, A Goode and D Chadwick (Eds.). Novartic Foundation, John Wiley & Sons Ltd., New York, Chichester, Brisbane, Singapore, Toronto, 242-251.

Chakravorty, A, P D Ghosh, P K Sahu, 2013. Multivariate analysis of phenotypic diversity of landraces of rice of West Bengal. *American Journal of Experimental agriculture*, 3(1): 110-123.

Chang, T T, G Loresto and O Tagumpay. 1974. Screening rice germplasm for drought resistance. *SABRAO Journal*, 6 (1): 9-16.

Clifford, S C, I M Stonach, C R Black, P R Singleton-Jones, S N Azam-Ali and N J M Crout. 2000. Effects of CO2 drought and temperature on the water relations and gas exchange of groundnut (*Arachis hypogaea*) stands grown in controlled environment glasshouses. *Physiologica plantarum*, 110: 78-88.

Clover, G R G, K W Jaggard, H G Smith and S N Azam-Ali. 2001. The use of radiation interception and transpiration to predict the yield of healthy, droughted and virus-infected sugar beet. *Journal of Agricultural Science*, 136: 169-178.

Collinson, S T, E J Clawson, S N Azam-Ali and C R Black. 1997. Effects of soil moisture deficits on the water relations of bambara groundnut (*Vigna subterranea* L. Verde.). *Journal of Experimental Botany*, 48 (309): 877-884.

Das, A and H Uchimiya. 2002. Oxygen stress and adaptation of a semi-aquatic plant: Rice (*Oryza sativa*). *Journal of Plant Research*, 115: 315–320.

Ekanayake, I J, J C O'Toole, D P Garrityand and T M Masajo. 1985. Inheritance of root characters and their relation to drought resistance in rice. *Crop Science*, 25: 927-933.

Farooq, M, N Kobayashi, A Wahid, O Ito and S M A Basra. 2009. Strategies for producing more rice with less water. *Advances in Agronomy*, 101: 351–388.

Farooq, M, N Kobayashi, O Ito, A Wahid and R Serraj. 2010. Broader leaves results in better performance of *indica* rice under drought stress. *Journal of Plant Physiology*, 167: 1066-1075.

Fukai, S and M Cooper. 1995. Development of drought resistant cultivars using physio-morphological traits in Rice. *Field Crop Research*, 40: 67-86.

Gupta, A, A Rico-Medina and A I Caño-Delgado. 2020. The physiology of plant responses to drought. *Science*, 368: 266–269.

Hao, Z C, V P Singh and Y L Xia. 2018. Seasonal drought prediction: Advances, challenges, and future prospects. *Reviews of Geophysics*, 56 (1): 108–141.

Hussain, H A, S Hussain, A Khaliq, U Ashraf, S A Anjum, S N Men and L C Wang. 2018. Chilling and drought stresses in crop plants: Implications, cross talk, and potential management opportunities. *Frontiers in Plant Science*, 9: 393.

Ingram, K T, J G Real, M A Maguling, M A Obien and G C Loresto. 1990. Comparison of selection indices to screen lowland rice for drought resistance. *Euphytica*, 48: 253-260.

IRRI (International Rice Research Institute). 1996. Standard Evaluation System for Rice. IRRI, Los Banos, Philippines. pp. 1-57.

Khalequzzaman, M. 2009. Development of drought resistant rice (*Oryza sativa* L.) varieties using in vitro hybridization. *Lambert Academic Publishing*, Germany: 135.

Khalequzzaman, M. 2002. Development of drought resistance varieties of Rice (*Oryza sativa* L.) through in vitro hybridization. PhD thesis, University of Southampton.

Kim, Y, Y S Chung, E Lee, P Tripathi, S Heo and K H Kim. 2020. Root response to drought stress in rice (*Oryza sativa* L.). *International Journal of Molecular Sciences*, 21 (4): 1513.

Kramer, P J. 1980. Drought, stress and the origin of adaptation. *In*: Adaptation of plants to water and high temperature stress, Turner, N C and P J Kramer (Eds.). *A willy interscience publication*, New York, Chichester, Brisbane, Toronto, 7-20.

Kumar, A, S Basu, V Ramegowda and A Pereira. 2016. Mechanisms of drought tolerance in rice. *In*: Achieving sustainable cultivation of rice, *Burleigh Dodds Science Publishing Limited*, UK. http://dx.doi.org/10.19103/AS.2106.0003.08.

Ludlow, M M and R C Muchow.1990. A critical evaluation of traits for improving crop yields in water-limited environments. *Advances in Agronomy*, 43: 107-153.

Melandri, G, H AbdElgawad, D Riewe, J A Hageman, H Asard, G T S Beemster, N Kadam, K Jagadish, T Altmann, C Ruyter-Spira and H Bouwmeester. 2020. Biomarkers for grain yield stability in rice under drought stress. *Journal of Experimental Botany*, 71 (2): 669–683.

Mishra, S S and D Panda. 2017. Leaf traits and antioxidant defense for drought tolerance during early growth stage in some popular traditional rice landraces from Koraput, India. *Rice Science*, 24 (4): 207–217.

Nelson, G C, H Valin H, R D Sands, P Havlík, H Ahammad H, D Deryng D, *et al.* 2014. Climate change effects on agriculture: Economic responses to biophysical shocks. *Proc. Natl. Acad. Sci. USA*, 111 (9): 3274–3279.

Oladosu, Y, M Y Rafii, C Samuel, A Fatai, U Magaji, I Kareem, Z S Kamarudin, I Muhammad and K Kolapo. 2019. Drought resistance in rice from conventional to molecular breeding: A review. *International Journal of Molecular Science*, 20 (14): 3519.

O'Toole, J C and M A Maguling. 1981. Greenhouse selection for drought resistance in rice. *Crop Science*, 21: 325-327.

O'Toole, J C. 1982. Adaptation of rice to drought prone environments. *In*: Drought resistance in crops with emphasis on rice. IRRI, Los Banos, Manilla, Philippines. pp. 195-216.

O'Toole, J C and T B Moya. 1978. Geotypic variation in maintenance of leaf water potential in rice. *Crop Science*, 18: 873-876.

Pantuwan, G, S Fukai, M Cooper, S Rajatasereekul and J C O'Toole. 2002. Yield response of rice (*Oryza sativa* L.) genotypes to different types of drought under rainfed lowlands Part 1. Grain yield and yield components. 2. Selection of drought resistant genotypes. 3. Plant factors contributing to drought resistance. *Field Crops Research*, 73: 153-200.

Panda, D, S S Mishra and P K Behera. 2021. Drought Tolerance in Rice: Focus on Recent Mechanisms and Approaches. *Rice Science*, 28 (2): 119-132. DOI: 10.1016/j.rsci.2021.01.002

Pandey, V and A Shukla. 2015. Acclimation and tolerance strategies of rice under drought stress. *Rice Science*, 22 (4): 147–161.

Ray, D K, N D Muller, P C West and J A Foley. 2013. Yield trends are insufficient to doubling global crop production by 2050. *PLoS One*, 8(6): e66428 doi:10.1371/jounal.pone.0066428.

Rollins J A, E Hab, S E Templer, T Colby, J Schmidt and M V Korff. 2013. Leaf proteome alterations in the context of physiological and morphological responses to drought and heat stress in barley (*Hordeum vulgare* L.). *Journal of Experimental Botany*, 64 (11): 3201–3212.

Sabouri, A, A R Dadras, M Azari, A S Kouchesfahani, M Taslimi and R Jalalifar. 2022. Screening of rice drought-tolerant lines by introducing a new composite selection index and competitive with multivariate methods. *Scientific Reports*, 12: 2163. https://doi.org/10.1038/s41598-022-06123-9.

Singh, R, Y Singh, S Xalaxo, S Verulkar, N Yadav,S Singh,N Singh, K S N Prasad, K Kondayya, P V Ramana Rao, M G Rani, T Anuradha, *et al.* 2016. From QTL to variety- harnessing the benefits of QTLs for drought, flood and salt tolerance in mega rice varieties of India through a multi-institutional network. *Plant Science*, 242: 278–287.

Thanh, N D, H G Zheng, N Y Dong, LN Trinh, M L Ali and H T Nguyen. 1999. Genetic variation in root morphology and microsatellite DNA loci in upland rice (*Oryza sativa* L.) from Vietnam. *Euphytica*, 105: 43-51.

Torres, R O, K L McNally, C V Cruz, R Serraj and A Henry. 2013. Screening of rice Genebank germplasm for yield and selection of new drought tolerance donors. *Field Crops Research*, 147: 12–22.

Turner, N C, G C Wright and K H M Siddique. 2001. Adaptation of grain legumes (Pulses) to water-limited environments. Advances in Agronomy, 71: 193-232.

Upadhyaya, H and S K Panda. 2019. Drought stress responses and its management in rice. *In*: Advances in Rice Research for Abiotic Stress Tolerance, M Hasanuzzaman, M Fujita, K Nahar, J K Biswas (Eds.). Woodhead Publishing, 177–200.

Vikram, P, A Kumar, A Singh and N K Singh. 2012. Rice: Genomics-assisted breeding for drought tolerance. *In*: Improving Crop Resistance to Abiotic Stress, N Tuteja, S S Gill, A F Tiburcio, and R Tuteja (Eds.). Wiley-VCH Verlag GmbH & Co. KGaA, 715–731.

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Genetic Diversity of INGER Rice Genotypes Based on Morphological Characters and Bacterial Blight Resistance

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T H Ansari[1] and M A Latif[1]

ABSTRACT

Bacterial blight disease (causal organism: *Xanthomonas oryzae* pv. *oryzae*) is an economically significant menace to rice cultivation in Bangladesh as well as in the world, which reduces significant yield loss in rice and hampers food security. The most sustainable strategy to fight this disease is the adoption of disease-resistant cultivars. The morphological trait and nature of diversity of 92 bacterial blight-resistant INGER (International Network for Genetic Evalu-ation of Rice) genotypes collected from the International Rice Research Institute (IRRI, Philippines) were analyzed to explore for sources of resistance. Artificial inoculation by *BXO9*, a virulent race of *Xoo* was used to evaluate and screen those genotypes in the field. Twelve genotypes, out of 92 had resistance, and another 12 had moderately resistance. Eleven morphological traits including disease data of each genotype were recorded and found noticeable diversity among the genotypes. Pearson's correlation analysis among genotypes revealed that yield per hill is positively correlated with number of tiller per hill, number of effective tiller per hill, number of spikelets per panicle, number of filled spikelets per panicle and thousand grain weight. In cluster analysis, 15 major groups were obtained in 92 rice genotypes by using Euclidean distance and the UPGMA method. Cluster-1 comprises single genotypes SVIN310, which showed resistant reaction to bacterial blight disease had the highest tiller number, effective tiller number, number of spikelets per panicle, filled spikelets per panicle and thousand-grain weight. In PCA analysis, the first four principal components narrated around 77.32% variation. Among 92 genotypes, G1 (SVIN310), G23 (SVIN018), G70 (SVIN012), G75 (SVIN054), G33 (SVIN007), G48 (SVIN006), G80 (SVIN049), G90 (BRRI dhan84), G30 (SVIN290) near to the vector line of yield per hill are highly and positively responsive to the yield per hill. Considering all of these, cluster-1 having genotype SVIN310 with resistant phenomena would be the potential genotype for further use in a breeding programme.

Key words: Bacterial blight, Disease resistance, Diversity analysis, INGER, Rice

INTRODUCTION

Rice (*Oryza sativa* L.), is the ancient domesticated and widely cultivated crop in the world (Ainsworth, 2008). From 2001 to 2025 the overall demand for rice will increase by 25% to bear the increasing population (Maclean *et al.*, 2002; Kabir *et al.*, 2020). In Bangladesh, rice security is equivalent to food security (Kabir *et al.*, 2020; Mamun *et al.*, 2021). Bangladesh which is recognized as one of the top climate vulnerable countries in the world, also facing the risk of climate change like severe drought, salinity, uneven precipitation, severe cold, and the emergence of diseases and pests (Mezanur-Rahman *et al.*, 2016; Mamun *et al.*, 2018; Rahman *et al.*, 2021; Aziz *et al.*, 2022). During the life cycle, rice faces different

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biotic and abiotic stresses like diseases, insects, submergence and salinity, etc. (Ara *et al.*, 2015; Morshed *et al.*, 2023). In Bangladesh, so far 32 diseases of rice is reported, and among them blast (Nihad *et al.*, 2022), bacterial blight (Rashid *et al.*, 2021), sheath blight (Latif *et al.*, 2022), false smut (Nessa *et al.*, 2015) and tungro (Nihad *et al.*, 2021; Hore *et al.*, 2022) diseases are the major threat of rice production. Enormous yield losses by rice diseases of bacterial, fungal, and viral hamper rice production (Ullah *et al.*, 2012). Bacterial Blight (BB) is one of the most disastrous rice diseases, which causes up to 50% yield loss in severe cases that are mostly dependent on variety, growth stage, geographical site, and ecological conditions (Liu *et al.*, 2014). Bacterial blight (BB) is first discovered in Fukuoka province, Japan in 1884 (Ou, 1985). Both inbred and hybrid varieties can be severely affected by BB disease and can cause significant yield loss (Anik *et al.*, 2022; Akter *et al.*, 2022). There is no doubt bacterial blight disease is a destructive disease, which can cause a serious problem and reduces yield in severe cases. Moreover, location wise variation of bacterial races makes it difficult to control and to date, 12 races of bacterial blight pathogen with diverse pathogenicity have been identified in Bangladesh (Rashid *et al.*, 2021). There are many different means of management like the use of some chemicals and antibiotics to control bacterial blight but it harms our environment and health. No effective chemical was found yet to give to the farmers for the management of BB in Bangladesh (Rahman *et al.*, 2018). Even, though bacterial blight is controlled by several measures, resistant variety is considered the durable and nature-friendly approach to control the disease (Nihad *et al.*, 2020; Akter *et al.*, 2022). Screening is the main gateway through which a breeder can identify the source of resistan genes and use them to develop durable disease-resistant rice varieties. According to, Anik *et al.*, 2022 it is the prerequisite to find out the potential resistant genotypes based on yield contributing morphological traits and nature of genetic diversity to develop a durable BB resistant variety. Thorough screening is obligatory to identify the resistant source from huge diverse populations. In plant breeding, genetic diversity plays a fundamental role to rescue resistant sources so breeders can develop stable variety after further screening and selection (Mazid *et al.*, 2013a; Nihad *et al.*, 2021). Researchers are always interested to identify a resistant cultivar to uncover available resistance genes against BB disease. It is reported that using gene pool, genome structure, and transferring desirable traits to plants is the most effective way for crop advancement (Nihad *et al.*, 2021; Anik *et al.*, 2022). Understanding and assessing genetic diversity is mandatory, which is the basis of plant breeding. A gene pool having diversified genetic resources is the prerequisite for initiating breeding programmes (Sivaranjani *et al.*, 2010; Nihad *et al.*, 2020). The objective of this experiment to evaluate the INGER rice genotypes against bacterial blight pathogen to find resistant sources against bacterial blight disease of Bangladesh.

MATERIALS AND METHODS

The experiment was set up in the research plot of BRRI, Gazipur during Boro 2018 following randomized complete block design (RCBD) with three replications. Ninety two rice germplasms (including resistant and susceptible checks) were obtained from International Rice Research Institute (IRRI) and Bangladesh Rice Research Institute (BRRI) to screen against bacterial blight disease. In every plot of each genotype, 15 plants were allowed to grow till harvesting. The plot size of each plot was 0.75m^2. Fertilizers were given in BRRI recommended doses and other intercultural practices were done in time as necessary. Five plants of each genotype were inoculated by a virulent race of *Xanthomonas oryzae* pv. *oryzae (Xoo)* by leaf clipping method at the maximum tillering stage.

Bacterial blight pathogen inoculation

A virulent *Xoo* isolate BXO9 (Khan *et al.*, 2009) was used for inoculation by following a well known leaf clipping method (Kauffman *et al.*, 1973). With an incubation period of 72 hours at 28°C, bacterial inoculum was prepared on Peptone Sucrose Agar (PSA) and mixed with distilled water for proper dilution. The suspension optical density (OD) absorbance was read at 600 nm and the concentration was adjusted to OD_{600} = 1. This value is equivalent to bacterial concentration of around 3.3×10^8 colony forming units per milliliter (cfu/mL). After dipping the scissors into the solution, about 3-4 cm healthy leaf portion from the top was cut for bacterial blight pathogen inoculation.

Data collection

Data of 11 morphological traits were documented from three hills of each genotype including disease reaction to bacterial blight disease. Plant height (PH, cm), number of tiller per hill (NTH^{-1} , no.), effective tiller per hill (ETH^{-1}, no.), days to 80% flowering (DF, day), days to maturity (DM, day), panicle length (PL, cm), number of grains per panicle (NGP^{-1}, no.), number of filled spikelet per panicle (FSP^{-1}, no), number of unfilled spikelet per panicle (USP^{-1}, no), thousand grains weight (TGW, g) and disease score (DS, no.) (Table 1) were the considerable traits for data collection. The disease data was collected at 14 days after inoculation from all leaves of three hills. Disease reaction was classified according to the standard evaluation scale of IRRI (IRRI, 2013) (Table 1). All susceptible, moderately susceptible and highly susceptible genotypes were considered as susceptible in this study.

Table 1. Disease scale for differentiate genotypes based on disease reaction.

Score	Disease Affected Leaf Area (%)	Description
1	1-5	Resistant
3	6-12	Moderately Resistant
5	13-25	Moderately Susceptible
7	26-50	Susceptible
9	>50	Highly Susceptible

Statistical analysis

Correlation, cluster and principal component analysis were done by using R programing software. Cluster analysis and dendrogram were prepared by using the Euclidean distance and UPGMA method. The principal coordinate analysis (PCoA) of 92 rice entries was done by EIGEN and PROJ modules of NTSYS-pc software.

RESULTS

Reaction of INGER genotypes to *Xoo*

Among 92 genotypes, 12 genotypes showed resistant, 12 showed moderately resistant and others genotypes showed susceptible reaction to bacterial blight disease (Table 2).

Table 2. List of INGER materials and disease reactions to *Xoo*.

Code	ID	Source of collection	Disease reaction	Code	ID	Source of collection	Disease reaction
G1	SVIN310	IRRI	R	G47	SVIN002	IRRI	S
G2	SVIN288	IRRI	R	G48	SVIN006	IRRI	S
G3	SVIN323	IRRI	R	G49	SVIN016	IRRI	S
G4	SVIN314	IRRI	R	G50	SVIN023	IRRI	S
G5	SVIN318	IRRI	R	G51	SVIN010	IRRI	S
G6	SVIN309	IRRI	R	G52	SVIN328	IRRI	S
G7	SVIN324	IRRI	R	G53	SVIN300	IRRI	S
G8	SVIN313	IRRI	R	G54	SVIN019	IRRI	S
G9	SVIN316	IRRI	R	G55	SVIN011	IRRI	S
G10	SVIN317	IRRI	R	G56	SVIN042	IRRI	S
G11	SVIN048	IRRI	MR	G57	SVIN030	IRRI	S
G12	SVIN312	IRRI	MR	G58	SVIN325	IRRI	S
G13	SVIN044	IRRI	MR	G59	SVIN013	IRRI	S
G14	SVIN005	IRRI	MR	G60	SVIN032	IRRI	S
G15	SVIN322	IRRI	MR	G61	SVIN043	IRRI	S
G16	SVIN045	IRRI	MR	G62	SVIN326	IRRI	S
G17	SVIN026	IRRI	MR	G63	SVIN329	IRRI	S
G18	SVIN305	IRRI	MR	G64	SVIN003	IRRI	S
G19	SVIN315	IRRI	MR	G65	SVIN304	IRRI	S
G20	SVIN307	IRRI	MR	G66	SVIN034	IRRI	S
G21	SVIN321	IRRI	MR	G67	SVIN035	IRRI	S
G22	SVIN050	IRRI	MR	G68	SVIN031	IRRI	S
G23	SVIN018	IRRI	S	G69	SVIN038	IRRI	S
G24	SVIN327	IRRI	S	G70	SVIN012	IRRI	S
G25	SVIN302	IRRI	S	G71	SVIN008	IRRI	S
G26	SVIN303	IRRI	S	G72	SVIN014	IRRI	S
G27	SVIN285	IRRI	S	G73	SVIN319	IRRI	S
G28	SVIN020	IRRI	S	G74	SVIN292	IRRI	S
G29	SVIN024	IRRI	S	G75	SVIN054	IRRI	S
G30	SVIN290	IRRI	S	G76	SVIN046	IRRI	S
G31	SVIN291	IRRI	S	G77	SVIN036	IRRI	S
G32	SVIN287	IRRI	S	G78	SVIN022	IRRI	S
G33	SVIN007	IRRI	S	G79	SVIN051	IRRI	S
G34	SVIN289	IRRI	S	G80	SVIN049	IRRI	S
G35	SVIN037	IRRI	S	G81	SVIN029	IRRI	S
G36	SVIN306	IRRI	S	G82	SVIN299	IRRI	S
G37	SVIN028	IRRI	S	G83	BRRI dhan28	BRRI	S
G38	SVIN009	IRRI	S	G84	BRRI dhan29	BRRI	S
G39	SVIN041	IRRI	S	G85	BRRI dhan50	BRRI	S
G40	SVIN039	IRRI	S	G86	BRRI dhan58	BRRI	S
G41	SVIN033	IRRI	S	G87	BRRI dhan63	BRRI	S
G42	SVIN021	IRRI	S	G88	BRRI dhan74	BRRI	S
G43	SVIN047	IRRI	S	G89	BRRI dhan81	BRRI	S
G44	SVIN004	IRRI	S	G90	BRRI dhan84	BRRI	S
G45	SVIN351	IRRI	S	G91	IRBB60	IRRI	R
G46	SVIN040	IRRI	S	G92	IRBB65	IRRI	R

R: Resistant, MR: Moderately Resistant, S: Susceptible, SVIN: Source of Variation INGER, G: Genotype.

Pearson's correlation coefficient

Correlation analysis revealed the relationship among the studied traits to take decision to design an effective breeding schedule. Effective tiller per hill had significant positive (0.92***) correlation with the total tiller per hill (Fig. 1). Additionally, number of filled spikelets (0.84***) as well as unfilled spikelets (0.67***) had positively related with total number of filled spikelets per panicle. From this study, it is showed that panicle length (0.59***) had positively correlated with plant height and yield per hill is positively correlated with number of spikelets per panicle (0.59***), number of filled spikelets (0.6***) and thousand grain weight (0.36***). Yield per hill also positively correlated with number of tiller per hill (0.6***) and number of effective tiller per hill (0.7***).

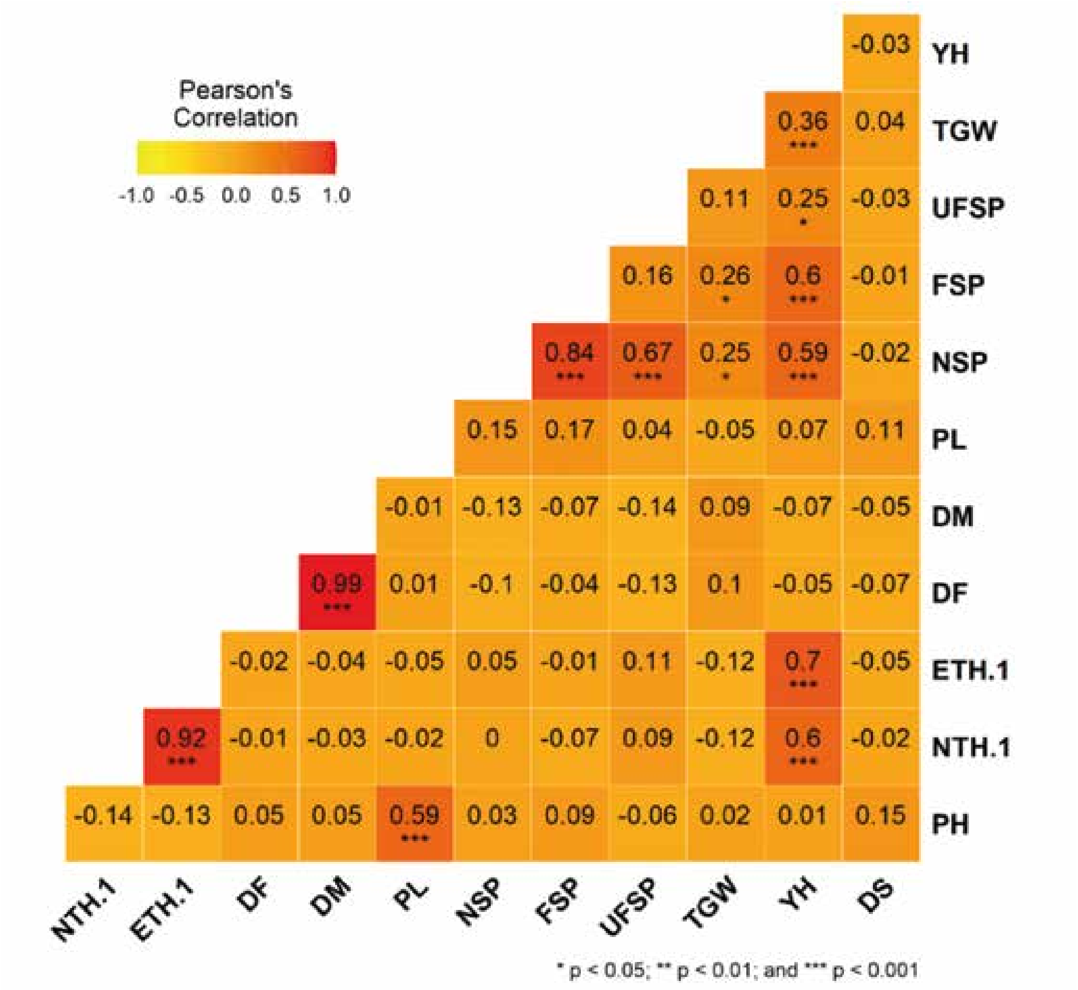


Fig. 1. Correlogram represented the relationship among the studied traits.
Here, PH: Plant height, NTH^{-1}: Number of tiller per hill, ETH^{-1}: Effective tiller per hill, DF: Days to 50% flowering, DM: Days to maturity, PL: Panicle length, NSP^{-1}: Number of spike-lets per panicle, FSP^{-1}: number of filled spikelets per panicle, UFSP^{-1}: Number of unfilled spikelets per panicle, TGW: Thousand grains weight, DS: Disease severity.

Cluster analysis

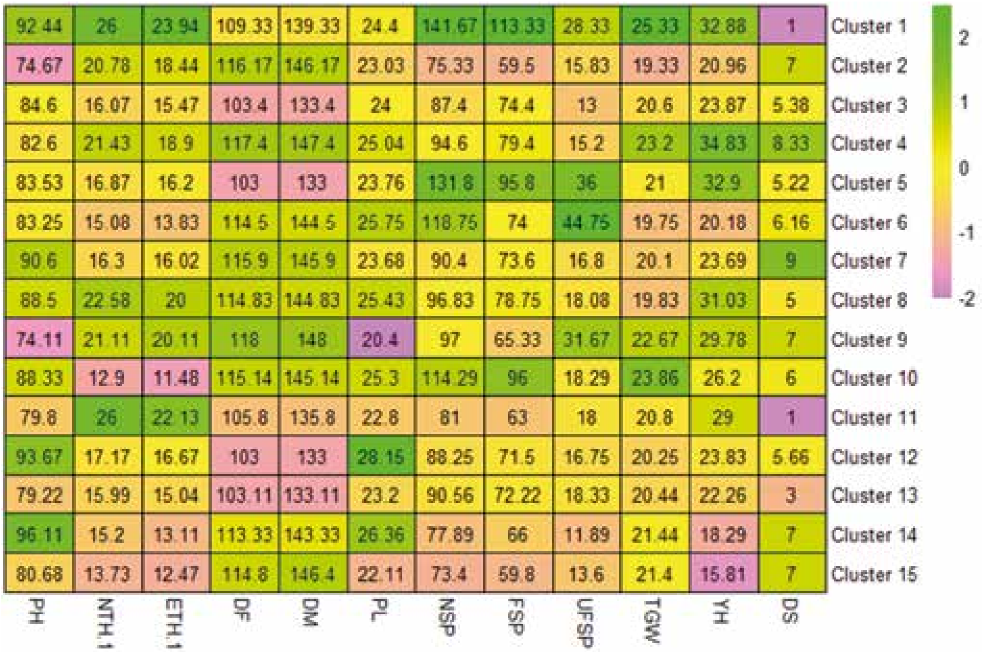
Based on multivariate analysis of morphological characters, 15 major groups were observed among 92 rice genotypes (Fig. 2 and Table 3). Cluster 3 had the highest number of genotypes (47), which comprised 51.08% of the studied genotypes. Clusters having the single genotype (clusters 1, 2, 7, 8, 9, 11, 13, 14 and 15) were considered the smaller groups compared to others. Clusters 10 was comprised of two genotypes, whereas, groups 4 and 12 containing three genotypes. On the other hand, the second largest cluster was group 6 as it comprised of 19 genotypes and cluster 5 containing nine genotypes.

Fig. 3 presents clusterwise mean data of studied parameters. Cluster-1 had the highest average number of tiller per hill, effective tiller per hill, number of spikelets per panicle, filled spikelets per panicle and thousand grain weight. Cluster-4 had the highest yield per hill. Cluster-11 had also the highest number of tiller per hill and cluster-12 had the highest number panicle length. Based on disease severity, cluster 1 and cluster 11 found as resistant to bacterial blight disease. Genotype of cluster 13 found as moderately resistant (Fig. 3).

Table 3. Number of cluster and respective genotypes found from Euclidean distance and UPGMA method cluster analysis.

Cluster	Genotype	Designation
1	G1	SVIN310
2	G70	SVIN012
3	G2, G3, G4, G6, G17, G18, G12, G13, G14, G16, G20, G21, G22, G23, G25, G27, G28, G35, G37, G38, G40, G41, G42, G44, G45, G46, G47, G49, G50, G53, G60, G64, G65, G72, G74, G75, G76, G79, G81, G85,G86, G87, G88, G89, G90, G91, G92	SVIN288, SVIN323, SVIN314, SVIN309, SVIN026, SVIN305, SVIN312, SVIN044, SVIN005, SVIN045, SVIN307, SVIN321, SVIN050, SVIN018, SVIN302, SVIN285, SVIN020, SVIN037, SVIN028, SVIN009, SVIN039, SVIN033, SVIN021, SVIN004, SVIN351, SVIN040, SVIN002, SVIN016, SVIN023, SVIN300, SVIN032, SVIN003, SVIN304, SVIN014, SVIN292, SVIN054, SVIN046, SVIN051, SVIN029, BRRI dhan50, BRRI dhan58, BRRI dhan63, BRRI dhan74, BRRI dhan81, BRRI dhan84, IRBB60, IRBB65
4	G24, G26, G36	SVIN327, SVIN303, SVIN306
5	G5, G8, G62, G71, G82, G78, G80, G30, G34	SVIN318, SVIN313, SVIN326, SVIN008, SVIN299, SVIN022, SVIN049, SVIN290, SVIN289
6	G9, G10, G31, G32, G66, G67, G68, G69, G51, G52, G54, G57, G59, G61, G63, G19, G77, G15, G29	SVIN316, SVIN317, SVIN291, SVIN287, SVIN034, SVIN035, SVIN031, SVIN038, SVIN010, SVIN322, SVIN019, SVIN030, SVIN013, SVIN043, SVIN329, SVIN315, SVIN036, SVIN328, SVIN024
7	G83	BRRI dhan28
8	G55	SVIN011
9	G84	BRRI dhan29
10	G43, G56	SVIN047, SVIN042
11	G7	SVIN324
12	G48, G33, G58	SVIN006, SVIN007, SVIN325
13	G11	SVIN048
14	G73	SVIN319
15	G39	SVIN041

Fig. 2. Circular dendrogram showing the cluster wise genotypes distribution of 92 rice genotypes.



PH	NTH.1	ETH.1	DF	DM	PL	NSP	FSP	UFSP	TGW	YH	DS	
92.44	26	23.94	109.33	139.33	24.4	141.67	113.33	28.33	25.33	32.88	1	Cluster 1
74.67	20.78	18.44	116.17	146.17	23.03	75.33	59.5	15.83	19.33	20.96	7	Cluster 2
84.6	16.07	15.47	103.4	133.4	24	87.4	74.4	13	20.6	23.87	5.38	Cluster 3
82.6	21.43	18.9	117.4	147.4	25.04	94.6	79.4	15.2	23.2	34.83	8.33	Cluster 4
83.53	16.87	16.2	103	133	23.76	131.8	95.8	36	21	32.9	5.22	Cluster 5
83.25	15.08	13.83	114.5	144.5	25.75	118.75	74	44.75	19.75	20.18	6.16	Cluster 6
90.6	16.3	16.02	115.9	145.9	23.68	90.4	73.6	16.8	20.1	23.69	9	Cluster 7
88.5	22.58	20	114.83	144.83	25.43	96.83	78.75	18.08	19.83	31.03	5	Cluster 8
74.11	21.11	20.11	118	148	20.4	97	65.33	31.67	22.67	29.78	7	Cluster 9
88.33	12.9	11.48	115.14	145.14	25.3	114.29	96	18.29	23.86	26.2	6	Cluster 10
79.8	26	22.13	105.8	135.8	22.8	81	63	18	20.8	29	1	Cluster 11
93.67	17.17	16.67	103	133	28.15	88.25	71.5	16.75	20.25	23.83	5.66	Cluster 12
79.22	15.99	15.04	103.11	133.11	23.2	90.56	72.22	18.33	20.44	22.26	3	Cluster 13
96.11	15.2	13.11	113.33	143.33	26.36	77.89	66	11.89	21.44	18.29	7	Cluster 14
80.68	13.73	12.47	114.8	146.4	22.11	73.4	59.8	13.6	21.4	15.81	7	Cluster 15

Fig. 3. Cluster wise mean of the studied parameters of 92 rice genotypes.

Here dark green indicates the highest value and dark purple indicates the lowest value, PH: Plant height, NTH^{-1}: number of tiller per hill, ETH^{-1}: effective tiller per hill, DF: days to 50% flowering, DM: days to maturity, PL: panicle length, NSP^{-1}: number of spikelets per panicle, FSP^{-1}: number of filled spikelets per panicle, UFSP^{-1}: number of unfilled spikelets per panicle, TGW: 1000-grain weight, DS: Disease severity.

Principal component analysis (PCA)

PCA biplot analysis revealed that variability of number of tiller and effective tiller per plant, number of grain per panicle and yield per hill were high in the 92 genotypes (Fig. 4). Yield contributing characters i.e., number of tiller and effective tiller per plant, number of grain per panicle, filled and unfilled grain and thousand grain weight are positively related with yield per hill of the genotypes. The genotypes G1 (SVIN310), G23 (SVIN018), G70 (SVIN012), G75 (SVIN054), G33 (SVIN007), G48 (SVIN006), G80 (SVIN049), G90 (BRRI dhan84), and G30 (SVIN290) that are close to the yield per hill vector line respond positively and highly to it. Resistant, moderately resistant and susceptible genotypes also showed the diversified position in PCA biplot analysis. First four principal components justified about 77.32% of the variability and showed a high correlation. The PC1, PC2, PC3 and PC4 described about 25.9%, 17.2%, 18.85 % and 15.37 % of the total variability (Table 4). In PC3, NTH^{-1} (0.72%), ETH^{-1} (0.72%), DF (0.63%) and DM (0.62%) were the most important contributing characters. On the other hand, NSP^{-1} (0.89%), FSP^{-1} (0.72%), $UFSP^{-1}$ (0.6%) is important parameters for the first PC. PH (0.81%) and PL (0.83%) is the most important trait for PC4 (Table 4).

Table 4. Eigen vectors and eigen values of the first four principal components.

Variable	Principal component			
	PC1	PC2	PC3	PC4
Eigen value	2.387	2.017	1.886	1.537
Percent	25.9	17.2	18.855	15.368
Cumulative	23.867	44.038	62.893	78.261
PH	0.103	0.283	0.017	0.819
NTH^{-1}	0.197	-0.606	0.722	0.123
ETH^{-1}	0.232	-0.601	0.721	0.111
DF	-0.485	0.575	0.634	-0.071
DM	-0.509	0.569	0.619	-0.081
PL	0.171	0.257	-0.013	0.836
NSP^{-1}	0.889	0.350	0.150	-0.142
FSP^{-1}	0.720	0.431	0.123	-0.098
$UFSP^{-1}$	0.609	0.021	0.100	-0.122
TGW	0.282	0.426	0.108	-0.290

Note. PH: Plant height, NTH^{-1}: number of tiller per hill, ETH^{-1}: effective tiller per hill, DF: days of 50% flowering, DM: days to maturity, PL: panicle length, NSP^{-1}: number of spikelets per panicle, FSP^{-1}: number of filled spikelets per panicle, $UFSP^{-1}$: number of unfilled spikelets per panicle, TGW: 1000-grain weight.

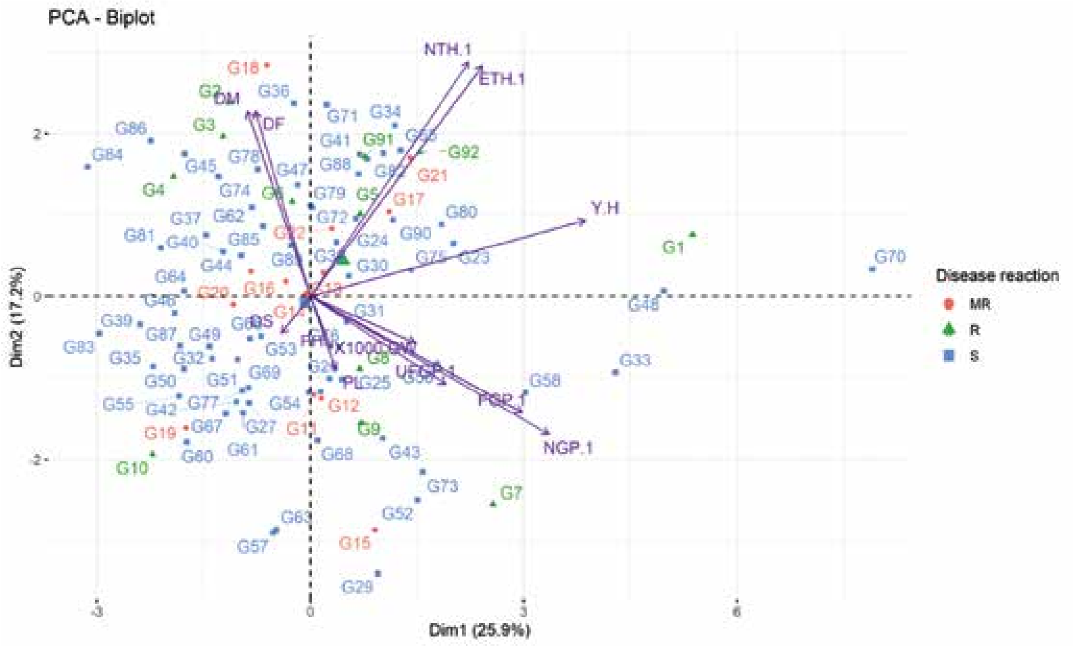


Fig. 4. PCA biplot of 92 rice genotypes based on morphological characters and disease reaction. Here, PH: Plant height, NTH^{-1}: number of tiller per hill, ETH^{-1}: effective tiller per hill, DF: days of 50% flowering, DM: days to maturity, PL: panicle length, NSP^{-1}: number of spikelets per panicle, FSP^{-1}: number of filled spikelets per panicle, UFSP^{-1}: number of unfilled spikelets per panicle, TGW: 1000-grain weight, DS: Disease severity.

Principal coordinate analysis (PCoA)

PCoA plot depicted the spatial dispersal of the genotypes (Fig. 5). SVIN319 (G73), SVIN012 (G70), BRRI dhan29 (G84), SVIN022 (G78), SVIN299 (G82) were found far away from center of the cluster. The rest of the genotypes were placed more or less near to the center (Fig. 5). In this case, center means that point where cluster center exists. In this point, at least one number for each parameter is present. Contour lines between each genotype and the center characterized Eigen vectors for the respective genotypes. The information generated from these results explained that genotypes those are far away from center are more genetically diverse and genotypes those are placed in near to the center are less diverse.

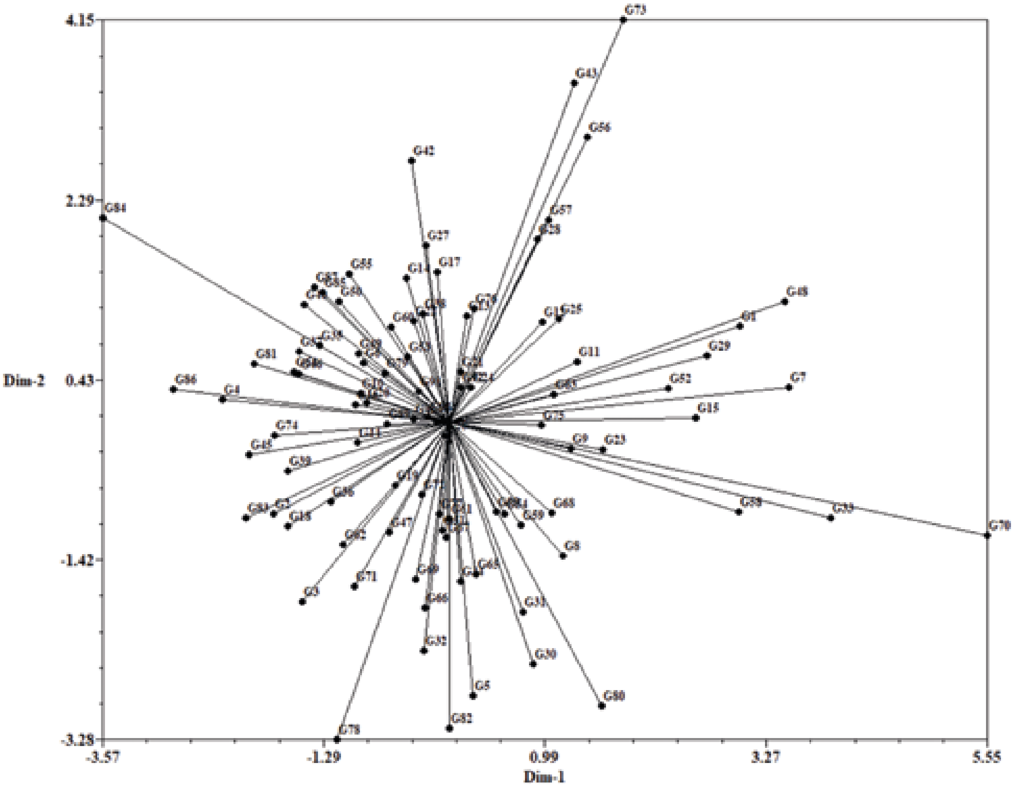


Fig. 5. Two-dimensional PCoA plot of 92 genotypes.

DISCUSSION

Revealing of diversity analysis based upon morphological traits and disease reaction are important to initiate resistant breeding programme. Yield is a quantitative trait and it regulates by so many factors. Indirect factors are plant height, growth duration, effective tiller number, length of panicle, seed length, seed setting and direct factors are panicle number, grains per panicle, filled grains and thousand grain weight (Sakamoto & Matsuoka, 2008; Huang *et al.*, 2011). Therefore, for improving yield related traits by direct selection sometimes become complicated and time demanding. Moreover, indirect selection is much easier and less time consuming. Consequently, it is suitable to use strongly correlated traits (Ahmadikhah *et al.*, 2008). Thousand grain weight is positively correlated with filled spikelets per panicle. All of those traits can contribute in enhancing yield of a genotype. Positive relationship between TGW and grain yield was described by Tsuzuki and Umeki, 1990. Furthermore, strong and significant relationship between yield and TGW also stated by the researchers (Mirza *et al.*, 1992; Efendi *et al.*, 2015). Significant relationship was found between filled spikelets and rice yield in this study, which corroborate with Ullah *et al.*, 2011. Additionally, panicle per hill positively correlated with rice yield which is similiar to the results of Abarshahr *et al.*, 2011. On the other hand, plant height has no significant relationship with the yield of the studied genotypes. Sarawgi *et al.*, 2013 described analogous results. Plant height and some other indirect traits have significantly weak relationship compared to the direct traits (Hairmansis *et al.*, 2013). Mohaddesi *et al.*, 2010 found that plant height and grain yield have a significant positive correlation.

By cluster analysis, 15 clusters were found from the distance analysis of the morphological traits of the studied genotypes. Based on 11 phenotypic traits 58 rice entries were grouped into four clusters reported by Ahmadikhah *et al.*, 2008. Based on 20 morphological traits, 23 rice lines were divided into ten different groups (Veasey *et al.*, 2008). The UPGMA dendrogram divided 41 bacterial blight resistant and susceptible rice varieties into six major clusters based on 13 agronomic traits (Mazid *et al.*, 2013b).

First four principal components of the present study described around 77.2% of variation. Lasalita Zapico *et al.*, 2010 also noted 82.7% of the total variability in 32 upland rice geno-types, which is almost similar to the results of our study. Eigenvectors specified the contribution of agronomic characters for percentage of variation to the principal components (Latif *et al.*, 2011). Moreover, 70.99% variability was described by four principal components derived from the analysis of 11 phenotypic traits of 94 rice entries (Nihad *et al.*, 2021). Caldo *et al.*, 2016 noted the first 10 principal components described for 67% of total variability of the agronomic traits.

Principal coordinate analysis display the spatial dispersal of the varieties based on their relatedness (Nihad *et al.*, 2021). Genotypes near to the centroid indicates they have similar characteristics, whereas genotypes distant from centroid indicates diverse characteristics (Nihad *et al.*, 2021). Siddique *et al.*, 2017 reported that the landraces distantly positioned from the center point were more diverse while the rice entries located near to the centroid carried more or less similar genetic composition and these findings support the result of the present study. Nevertheless, genotypes having broader deviation could be utilized as donor parents for hybridization to advance bacterial blight resistant variety.

CONCLUSION

Information generated from this study might be helpful for breeders to select resistant materials considering yield contributing character for durable bacterial blight resistant variety development. In Pearson's correlation coefficient, it is showed that yield per hill is positively correlated with number of spikelets per panicle (0.59***), number of filled spikelet (0.6***), thousand grain weight (0.36***), number of tiller per hill (0.6***) and number of effective tiller per hill (0.7***). Cluster 1 comprising single genotypes (SVIN310) showed the highest number of tiller, effective tiller, number of filled spiklets per panicle. PCA biplot analysis revealed that variability of number of tiller and effective tiller per plant, number of spikelets per panicle and yield per hill were high among the 92 genotypes. Yield contributing characters i.e., number of tiller and effective tiller per plant, number of spikelets per panicle, filled spikelets per panicle and thousand grain weight are positively related with yield per hill of the genotypes. Out of 92 INGER genotypes G1 (SVIN310), G23 (SVIN018), G70 (SVIN012), G75 (SVIN054), G33 (SVIN007), G48 (SVIN006), G80 (SVIN049), G90 (BRRI dhan84), G30 (SVIN290) near to the vector line of yield per hill are highly and positively responsive to the yield per hill. The mentioned genotypes could be used in hybridization programme to develop high yielding variety. In another words, the entry G1 (SVIN310) which have both yield potential and bacterial blight resistance phenomena could be used as resistant source to develop bacterial blight resistant variety.

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REFERENCES

Ara, A, K M Iftekharuddaula, M M H Saikat, A B M A Uddin and M A I Khan. 2015. Introgression of *Sub1* QTL into a rainfed lowland rice variety of Bangladesh using marker assisted backcross approach. *International Journal of Research*, 2 (5): 2348-4.

Abarshahr, M, B Rabiei, H Samizadeh Lahigi. 2011. Assessing genetic diversity of rice varieties under drought stress conditions. *Notulae Scientia Biologicae*. 3: 114–123. https://doi.org/10.15835/nsb315618.

Ahmadikhah, A, S Nasrollanejad and O Alishah. 2008. Quantitative studies for investigating variation and its effect on heterosis of rice. *International Journal of Plant Production*. https://doi.org/10.22069/ijpp.2012.621.

Ainsworth, E A. 2008. Rice production in a changing climate: a meta-analysis of responses to elevated carbon dioxide and elevated ozone concentration. *Global Change Biology*. 14: 1642–1650. https://doi.org/10.1111/J.1365-2486.2008.01594.X.

Akter, A, S A I Nihad, M J Hasan, L Hassan, A H K Robin, M R Quddus, A Tabassum and M A Latif. 2022. Evaluation of hybrid rice parental lines against bacterial blight disease and detection of resistant gene (s) by gene-specific, linked markers. *Journal of Phytopathology*, 170: 382–390. https://doi.org/10.1111/JPH.13087.

Anik, T R, S A I Nihad, M A I Hasan, M A Hossain, M M Rashid, M A I Khan, K P Halder and M A Latif. 2022. Exploring of bacterial blight resistance in landraces and mining of resistant gene(s) using molecular markers and pathogenicity approach. *Physiology and Molecular Biology of Plants*. 282 (28): 455–469. https://doi.org/10.1007/S12298-022-01139-X.

Aziz, M A, H U S Shohan, N M F Rahman, M C Rahman, S A I Nihad, S M Q Hassan, M S Kabir, M I Hossain, R Ahmed, M A Qayum, M A Al Mamun, F Rahman and Z Rukshanara. 2022. Projection of future precipitation in Bangladesh at Kharif-II season using geospatial techniques. *Earth Systems and Environment*, 2022: 1–12. https://doi.org/10.1007/S41748-022-00319-9.

Caldo, R, L Sebastian, and J Hernandez, 1996. Morphology based genetic diversity analysis of ancestral lines of Philippine rice cultivars. *Philippine Journal of Crop Science*. 21(3): 82-96.

Efendi, Kesumawaty, S Zakaria, Bakhtiar and Syafruddin. 2015. Morpho agronomic performance of rice (*Oryza sativa* L.) landraces under organic cultivation of SRI methods. *International Journal of Agricultural Research*, 10 (2):74-82.

Hairmansis, A, A Hairmansis, B Kustianto, S Suwarno. 2013. Correlation analysis of agronomic characters and grain yield of rice for tidal swamp areas. *Indonesian Journal of Agricultural Science*, 11: 11–15. https://doi.org/10.21082/ijas.v11n1.2010.p11-15.

Hore, T K, M A Inabangan-Asilo, R Wulandari, M A Latif, S A I Nihad, J E Hernandez, G B Gregorio, T U Dalisay, M Genaleen, Q Diaz, B Ch and B P M Swamy. 2022. Introgression of tsv1 improves tungro disease resistance of a rice variety BRRI dhan71. *Scientific Reports*, 12:1–14. https://doi.org/10.1038/s41598-022-23413-4

Huang, M, Y. bin Zou, P Jiang, B Xia, I Md Ao and H jun. 2011. Relationship between grain yield and yield components in super hybrid rice. *Agricultural Sciences in China*, 10: 1537–1544. https://doi.org/10.1016/S1671-2927(11) 60149-1.

IRRI. 2013.http://www.knowledgebank.irri.org/ricebreedingcourse/bodydefault.htm#Breeding for disease resistance Blight.htm.

Kabir, M S, M Salam, A Islam, M A R Sarkar, M Mamun, M C Rahman, B Nessa, M Kabir, H Shozib, M B Hossain, A C howdhury, M Nasim, K Iftekharuddaula, M S Hossain, M Bhuiyan, B Karmakar, M S Rahman, M Haque, M Khatun, M Ali, S Rabbi, P Biswas, E Rashid and N Rahman. 2020. Doubling rice productivity in Bangladesh: A way to achieving SDG 2 and moving forward. *Bangladesh Rice Journal*, 24: 1–47. https://doi.org/10.3329/BRJ.V24I2.53447.

Kauffman, H, A Reddy, S Hsieh and S Merca. 1973. An improved technique for evaluating resistance of rice varieties to *Xanthomonas oryzae*. Plant Dis Report.

Khan, M A I, M S Kabir, M A Monsur, M A Ali and M A T Mia. 2009. Pathogenic diversity of *Xanthomonas oryzae* pv. oryzae in Bangladesh. *Bangladesh Journal of Plant Pathology*, 25: 1–6.

Lasalita-Zapico, F C, J A Namocatcat and J L Carini-Turner. 2010. Genetic diversity analysis of traditional upland rice cultivars in Kihan, Malapatan, Sarangani Province, Philippines using morphometric markers. *The Philippine Journal of Science*, 139 (2): 177-180.

Latif, M A, S A I Nihad, M S Mian, S Akter, M A I Khan and M A Ali. 2022. Interaction among sheath diseases complex of rice and ribosomal DNA analysis for the differentiation of *Rhizoctonia solani*, R. oryzae and R. *oryzae-sativae*. *Plant Stress 5*, 100100. https://doi.org/https://doi.org/10.1016/j.stress.2022.100100

Latif, M A, M Rafii Yusop, M Motiur Rahman and M R Bashar Talukdar. 2011. Microsatellite and minisatellite markers based DNA fingerprinting and genetic diversity of blast and ufra resistant genotypes. *Comptes Rendus - Biologies*. https://doi.org/10.1016/j.crvi.2011.02.003.

Liu, W, J Liu, L Triplett, J E Leach and G L Wang. 2014. Novel insights into rice innate immunity against bacterial and fungal pathogens. *Annual review of phytopathology*, 52: 213–241. https://doi.org/10.1146/ANNUREV-PHYTO-102313-045926

Maclean, J L, D C Dawe, B Hardy and G P Hettel. 2002. Rice Almanak. Research Quarterly, 27 (1993): pp. 71-80.

Mamun, A Al, M N F Rahman, M Abdullah Aziz, M A Qayum, M I Hossain, S A I Nihad and M S Kabir. 2018. Identification of meteorological drought prone area in Bangladesh using Standardized Precipitation Index. *Journal of Earth Science & Climatic Change*. 09. https://doi.org/10.4172/2157-7617.1000457

Mamun, M A Al, S A I Nihad, M A R Sarkar, M A Aziz, M A Qayum, R Ahmed, N M F Rahman, M I Hossain and M S Kabir. 2021. Growth and trend analysis of area, production and yield of rice: A scenario of rice security in Bangladesh. PLoS One 16, e0261128. https://doi.org/10.1371/JOURNAL.PONE.0261128.

Mazid, S Muhammad, M Y Rafii, M M Hanafi, H A Rahim and M A Latif. 2013a. Genetic variation, heritability, divergence and biomass accumulation of rice genotypes resistant to bacterial blight revealed by quantitative traits and ISSR markers. Physiologia Plantarum. https://doi.org/10.1111/ppl.12054.

Mazid, M S, M Y Rafii, M M Hanafi, H A Rahim, M Shabanimofrad and M A Latif. 2013b. Agro-morphological characterization and assessment of variability, heritability, genetic advance and divergence in bacterial blight resistant rice genotypes. *South African Journal of Botany*. https://doi.org/10.1016/j.sajb.2013.01.004.

Mezanur-Rahman, M, M Anamul-Haque, S Arafat-Islam-Nihad, M Mahmudul-Hasan-Akand and M Ruhul-Amin-Howlader. 2016. Morpho-physiological response of *Acacia auriculiformis* as influenced by seawater induced salinity stress. Forest Systems. 25, e071. https://doi.org/10.5424/fs/2016253-09386.

Mirza, M J, F A Faiz and A Mazid. 1992. Correlation studies and path analysis of plant height yield and yield components in rice (*Oryza sativa* L.). *Sarhad Journal of Agricultural*, 8: 647-653.

Mohaddesi, A, A Abbasian, S Bakhshipoor and S M Mohammad. 2010. Study of effects of nitrogen fertilizer and planting distance on yield and yield components of promising rice line. *Journal of Crop Ecophysiology*, 2(3): 198-203.

Morshed, M N, M A A Mamun, S A I Nihad, M M Rahman, N Sultana, M M Rahman. 2023. Effect of weather variables on seasonal abundance of rice insects in southeast coastal region of Bangladesh. *Journal of Agriculture and Food Research*, 11, 100513. https://doi.org/10.1016/j.jafr.2023.100513

Nessa, B, M U Salam, A H M M Haque, J K Biswas, M A Latif, M A Ali, T H Ansari, M Ahmed, N Parvin, M Zakaria, I Baki, S Islam, M S Islam and J Galloway. 2015. Rice false smut disease at different flowering times. *Bangladesh Rice Journal*, 19: 28–35. https://doi.org/10.3329/BRJ.V19I2.28162.

Nihad, S A I, A Ara, M Rashid, M Hasan, M Khan and M Latif. 2020. Genetic divergence of rice genotypes revealed by bacterial blight disease and morphological traits. *Bangladesh Rice Journal*. https://doi.org/10.3329/brj.v24i1.53241.

Nihad, S A I, M K Hasan, A Kabir, M A I Hasan, M R Bhuiyan, M R Yusop and M A Latif. 2022. Linkage of SSR markers with rice blast resistance and development of partial resistant advanced lines of rice (*Oryza sativa*) through marker-assisted selection. *Physiology and Molecular Biology of Plants*, 281(28):153–169. https://doi.org/10.1007/s12298-022-01141-3

Nihad, S A I , A C Manidas, K Hasan, M A I Hasan, O Honey and M A Latif. 2021. Genetic variability, heritability, genetic advance and phylogenetic relationship between rice tungro virus resistant and susceptible genotypes revealed by morphological traits and SSR markers. *Current Plant Biology*. 25, 100194. https://doi.org/10.1016/j.cpb.2020.100194

Ou, SH. 1985. Rice Diseases. Commonwealth mycological institute, Kew, England.

Rahman, J R, M M Rashid, S A I Nihad, A Ara, M R Islam and M A I Khan. 2018. Evaluation of chemicals against bacterial blight of rice caused by *Xanthomonas oryzae* pv *oryzae*. *Bangladesh Journal of Agriculture*, 41–43: 1–12.

Rahman, M, H Rashid, M Kamal Shahadat, A A Topu, A Hossain, S A I Nihad, 2021. Field performance of some potato varieties under different saline conditions of Bangladesh. African Journal of Agricultural Research, 17: 1480–1487. https://doi.org/10.5897/AJAR2021.15578.

Rashid, M M, S A I Nihad, M A I Khan, A Haque, A Ara, T Ferdous, M.A Hasan and M.A Latif. 2021. Pathotype profiling, distribution and virulence analysis of *Xanthomonas oryzae* pv. *oryzae* causing bacterial blight disease of rice in Bangladesh. *Journal of Phytopathology*. 169: 438–446. https://doi.org/10.1111/jph.13000.

Sakamoto, T and M Matsuoka. 2008. Identifying and exploiting grain yield genes in rice. *Current opinion in plant biology*. 11, 209–214. https://doi.org/10.1016/J.PBI.2008.01. 009.

Sarawgi, A K, R L V Subba, M Parikh, B Sharma and G C Ojha. 2013. Assessment of variability of rice (*Oryza sativa* L.) germplasm using morphological characters. *Journal of Rice Research*. 6(1): 14-28.

Siddique, M., M Khalequzzaman, K Fatema, M. Islam, M Islam, & M Chowdhury. 2017. Molecular characterization and genetic diversity of aman rice (*Oryza sativa* L.) landraces in Bangladesh. *Bangladesh Rice Journal*, 20(2), 1–11. https://doi.org/10.3329/brj.v20i2.34123

Sivaranjani, A K P, M K Pandey, I Sudharshan, G R Kumar, M S Madhav, R M Sundaram, G S Varaprasad, N S Rani. 2010. Assessment of genetic diversity among basmati and non-basmati aromatic rices of India using SSR markers. *Current Science*, 99: 221–226.

Tsuzuki, E and Y Umeki. 1990. Studies on the methods of stabilizing and increasing yield in early-cultivated rice, correlation between grain yield and characters related with yield. Bulletin faculty Agriculture, Miyazaki University, Japan, 36, pp. 261-269.

Ullah, I, S Jamil, M Z Iqbal, H L Shaheen, S M Hasni, S Jabeen, A Mehmood, M Akhter, 2012. Detection of bacterial blight resistance genes in basmati rice landraces. *Genetics and molecular research: GMR*. 11, 1960–1966. https://doi.org/10.4238/2012.JULY.20.1

Ullah, M Z, M K Bashar, M S R Bhuiyan, M Khalequzzaman and M J Hasan. 2011. Interrelationship and cause-effect analysis among morpho-physiological traits in biroin rice of Bangladesh. *International Journal of Plant Breeding and Genetics*. https://doi.org/10.3923/ijpbg.2011.246.254.

Veasey, E A, E F Da Silva, E A Schammass, G C X Oliveira and A Ando. 2008. Morphoagronomic genetic diversity in american wild rice species. *Brazilian Archives of Biology and Technology*. 51, 94–104. https://doi.org/10.1590/S1516-89132008000100012.

Bangladesh Rice J.26 (1): 33-46, 2022, doi.org/10.3329/brj.v26i1.66591

Recent Advances in Population Improvement through RGA under Irrigated Boro Rice Breeding Programme in Bangladesh

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ABSTRACT

Development of new high yielding varieties needs highly accelerated breeding approaches to meet the demand of increased rice production nowadays. To address this issue, Rapid Generation Advance (RGA) of segregating rice population and Line Stage Testing (LST) of fixed breeding lines have been adopted as a routine work in Bangladesh Rice Research Institute. In the present study, we described the evidence of advancing a large number of segregating population and high selection pressure at LST to isolate fixed lines in the Irrigated Boro rice breeding programme. This programme was implemented under development of Favourable Boro Rice and Cold Tolerant Rice. In both programmes, wide variation was observed among the number of population and segregating generation at RGA nursery. A total of 62,269 individual progenies from 64 crosses were advanced through RGA in 2019 whereas 12,122 individual progenies from 45 crosses were advanced in 2020 under the FBR programme. In contrast, 68,531 segregating progenies from 82 crosses and 15,004 progenies from 52 crosses were advanced through RGA under CTR programme during 2019 and 2020, respectively. During LST, selection pressure was imposed for the first time to isolate homozygous lines from a pool of large number of breeding lines. The selection intensity ranged between 0.21-20.1% and 0.6-14.9% for FBR and CTR, respectively. A total of 794 fixed breeding lines having different combinations of favourable alleles of BLB, Blast and different grain quality traits were isolated from 17,633 RGA derived lines. The results obtained from this study suggested that the RGA system has become an effective tool for population improvement in a breeding programme.

Key words: Rapid generation advance, Line Stage Testing, breeding population, trait markers

INTRODUCTION

Rice grows round the year but traditionally there are three rice growing seasons in Bangladesh. Among these seasons, *Boro*, which corresponds to the irrigated ecosystem in the dry season (Afrin *et al.*, 2019) solely occupies around 45% of the rice areas and produces around 55% of rice annually (BBS, 2018). The rice varieties developed and released by the Bangladesh Rice Research Institute (BRRI) for the irrigated ecosystem have made a huge contribution to national food security. Around 70% of the total rice produced in the country in Boro season comes from BRRI varieties. However, only two varieties, BRRI dhan28 and BRRI dhan29 are widely grown covering over 50% of the rice areas in Boro season (Ahmed *et al.*, 2022). These varieties are very old and are showing weakness to different biotic and abiotic stresses. Therefore, development of high yielding farmers' adapted varieties for the irrigated ecosystem is urgently needed.

The rice varieties released so far were developed through the pedigree breeding method, which usually takes longer time (typically 6-8 years) to isolate a superior line from the segregating generations and

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eventually requires 12-14 years to release a new variety. Pedigree based breeding method has intrinsic weakness in improving the quantitatively inherited trait like yield as it favours major effect genes and takes longer time to recycle the progenies in the crosses. Thus, faster improvement of varieties for the favourable ecosystem breaking present yield plateau has been almost impossible through pedigree method. This method allows early generation selection from segregating population, which does not guarantee of producing high yielding progenies in the late generations (Collard *et al*., 2017). The reason behind this is the continuous decay of linkage disequilibrium among the small effect multiple genes/QTLs conferring yield because of breeders' favour for major genes during early generation selection. Besides, population size is a prime factor for capturing variations of small effect genes/QTLs (Cobb *et al.* 2019a). In pedigree method, although the population size in the early generation (F_2) remains relatively larger (2000 – 3000 progenies), due to selection pressure at successive generations, it makes low chance of capturing the small effects genes/QTLs from the respective crossing population. Moreover, there is a common tendency of the plant breeders to include landrace varieties frequently in the breeding programme to enrich specific traits of current demand, which in turn decays the elite status of the breeding germplasm (Cobb *et al*., 2019b) and lowers the yield potential. Also, heavily dependence on a few high yielding genetic backgrounds as parents in the breeding programmes has resulted into below average progenies. In contrast, rapid generation advance (RGA), a cross-cutting technology, has come up to speed up the breeding programme further to shorten the breeding cycle (Goulden, 1939, Fuente *et al*., 2013). In RGA methods, progeny selection is not practiced during advancement of segregating generations; rather successive planting following single seed descent reduces the time for line fixation. This system not only shortens the cycle time but also provides scope of capturing variations of small effects genes/QTLs for yield (Cobb *et al*., 2019b). Considering an increased demand for food, climate vulnerability and emerging biotic stresses, rice breeders need to adopt quicker and effective breeding operations to reduce the timeframe required for a complete breeding pipeline (Atlin *et al*., 2017; Collard *et al*., 2019).

The segregating populations grown at a closer spacing in the RGA system are forced to compete with the neighbouring plants for water, air and sunlight. As the principles of nature, this imposed competition leads the plants to flower earlier than the plants grown under standard spacing used for crop cultivation. This makes possible to advance 3-4 generations per year in rice. Segregating generations could be advanced without selection and in the successive generations (Goulden, 1939). Additionally, RGA has been proved as an efficient method to fix transgressive segregation and obtain genetic advance (Snape and Riggs, 1975). However, advancement of segregating population has created immense opportunity to develop and advance large scale population improvement in cereal crops. In recent times, RGA techniques have been adapted in different breeding programmes of different crops, viz, soybeans (Carandang *et al*., 2006), pigeon pea (Saxena *et al*., 2019), chickpea (Gaur *et al*., 2008), sorghum (Rizal *et al*., 2014), rice (Ohnishi *et al*., 2011; Tanaka *et al*., 2016; Collard *et al*., 2017; Rahman *et al*., 2019) in developing fixed breeding lines within possible shorter time. More recently, seven generations of oats and triticale (Liu *et al*., 2016) had been advanced in one year through RGA. Watson *et al*., 2018 achieved six generations per year in wheat, barley and chickpea.

The use of RGA in the breeding programme is not new, several varieties in different crops have been developed using this technique (Collard *et al.*, 2017). RGA is now in routine use in many rice breeding programmes in Asia including Bangladesh. Since 2015, BRRI has been using RGA technique in the breeding programmes for different ecosystem including the irrigated rice favourable ecosystems. In this article, we describe our experience and outcomes of using RGA techniques in shortening breeding cycle and isolating potential candidates from early generation multi-location trials of RGA-derived fixed lines.

MATERIALS AND METHODS

Plant materials

Segregating population of different crosses and RGA-derived fixed lines developed by the breeding programme for irrigated ecosystem were used in this study.

Line fixation through RGA techniques

Segregating (F_2 to F_6) populations were grown in the field RGA nursery (Fig 1). A portion of a single panicle having 8-10 seeds was sown at 5 X 5 cm spacing on the raised bed of 50 cm width. A wooden frame was used to make furrows at 5 cm apart on the fertilized and puddled bed to facilitate seeding. Gravel-free powdered garden soil was used to cover the seeds sown in the furrow. Fertilizer management during final land preparation was done with Di-Ammonium Phosphate (DAP), Muriate of Potash, Gypsum and Zinc Sulphate at the rate of 45, 50, 55 and 5.5 kg/ha, respectively. Besides, 50 kg Urea and 25 kg Muriate of Potash were applied at 40-45 days. At maturity, a secondary branch of a single panicle from each hill was harvested and dried. Dried seeds were immediately put into 50°C for 72 hours in oven for breaking the dormancy and proceed for the next cycle of RGA.

Seed amplification of the fixed lines

RGA derived fixed lines were grown in 2.4 m (12 hills) long single-row plots with 20 cm × 20 cm spacing following systematic arrangement design in a trial called Line-Stage Testing (LST). Forty-five-day-old seedlings were transplanted using single seedling per hill. At flowering stage, superior and uniform lines were selected based on days to 50% flowering, plant height, grain type. Lodging tolerance, reaction to major diseases and insects under field condition were also considered for line selection.

Genotyping of RGA derived fixed lines

Genotyping was performed through outsourcing at the Intertek, Australia with the help of International Rice Research Institute (IRRI). A leaf sample of 4-5 cm was collected in glassine bags from a single plant of each of the selected uniform lines of the LST trial and stored at -80°C until it was fridge dried using a lyophylizer machine. Two small, dried leaf discs from each sample were taken into each well of a 96-well plate using a paper punching machine. The 96 well-plates with leaf samples were kept in the oven at 60°C for two days followed by one day cooling period in room temperature before sending to the Intertek, Australia for genotyping. Trait SNP panel designed by the GSL laboratory of IRRI was used for genotyping.

RESULTS

Segregating population in the RGA nursery

The Favourable Boro Rice (FBR) and Cold Tolerant Rice (CTR) Breeding programme of BRRI advances F_2 –F_6 segregating progenies in the Field RGA nurseries with 2.5 generations a year. This breeding programme had advanced a sum of 62,269 progenies from 64 crosses and 68,531 segregating progenies from 82 crosses in 2019 in order to develop breeding lines suitable for favourable and cold prone ecosystem, respectively. Sequentially, in 2020, 12,122 progenies of favourable ecosystem and 15,004 progenies for cold prone environment were advanced (Table 1). In 2019, majority of the segregating progenies (~ 50%) were under F_2 class in both of the breeding programmes. Under favourable Boro rice (FBR) programme 32,202 F_2 progenies from 26 crosses, 13,303 F_4 progenies from nine crosses, 8,388 F_5 progenies from 12 crosses, and 8,376 F_6 progenies from 17 crosses were advanced in 2019, while in 2020, 3,096 F_2 progenies from 10 crosses, 7,349 F_4 progenies from 26 crosses and 1,777 F_6 progenies from 9 crosses were advanced. In contrast, in the cold tolerant rice (CTR) breeding programme, a total of 29,095 F_2 progenies from 33 crosses, 10,859 F_3 progenies from nine crosses, 10,077 F_4 progenies from five crosses, 12,544 F_5 progenies from 13 crosses, and 5,956 F_6 progenies from 22 crosses in 2019 and 1,048 F_2 progenies from five crosses, 8,066 F_4 progenies from 33 crosses, 3,296 F_5 progenies from nine crosses and 1,994 F_6 progenies from five crosses were advanced in 2020.

Table 1. The number of segregating progenies advanced by field RGA nurseries under FBR and CTR breeding programme in 2019 and 2020.

programme	Year					
	2019			2020		
	Generation	**Crosses**	**Population**	**Generation**	**Crosses**	**Population**
FBR	F_2	26	32,202	F_2	10	3,096
	F_4	9	13,303	F_4	26	7,349
	F_5	12	8,388	F_5	-	-
	F_6	17	8,376	F_6	9	1,777
	Total	**64**	**62,269**		**45**	**12,222**
CTR	F_2	33	29,095	F_2	5	1,048
	F_3	9	10,859	F_3	-	-
	F_4	5	10,077	F_4	33	8,666
	F_5	13	12,544	F_5	9	3,296
	F_6	22	5,956	F_6	5	1,994
	Total	**82**	**68,531**		**52**	**15,004**

Population structure of the segregating progenies

The population structure of the segregating progenies derived from RGA showed wide variations across the segregating population (F_2-F_6) under both FBR and CTR breeding programme. In 2019, the number of progenies per crosses in FBR breeding programme ranged from 442-1912 for F_2, 1325-1745 for F_4, 302-1154 for F_5, and 33-1060 for F_6 (Fig. 1). In CTR, higher number of crosses was in F_2 generation comprising 29,095 progenies with wide range of variation in number of progenies per crosses. The F_2 generations under CTR breeding programme had 182-1889 lines per cross (Fig. 2).

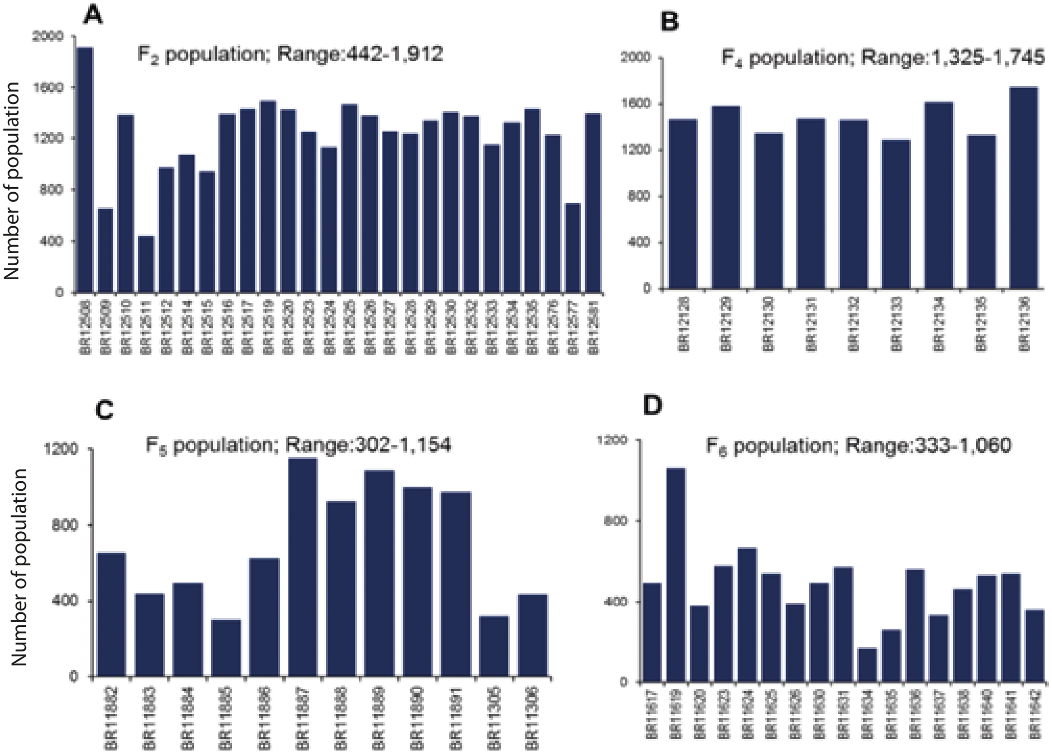

Fig. 1. Histogram showing number of populations produced in F_2 generation (A), F_4 generation (B), F_5 generation (C), and F_6 generation (D) under field RGA nursery of FBR programme during Boro 2019.

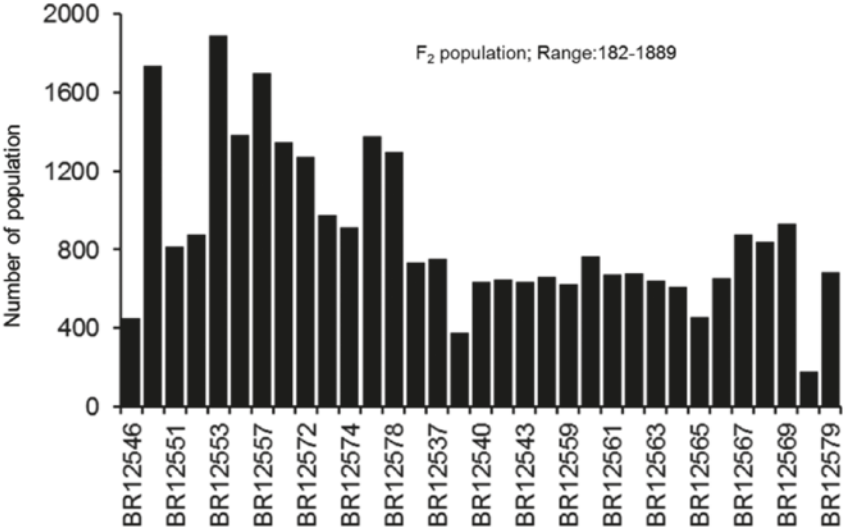

Fig. 2. Bar diagram showing number of population per crosses produced in F_2 generation field RGA nursery of CTR during Boro 2018-19.

In case of F_3-F_6 population, greater variations were observed for each progeny. The number of populations varied from 182-1889 for F_2, 913-1712 for F_3, 1457-2364 for F_4, 553-1386 for F_5, and 29-690 for F_6 (Fig. 3).

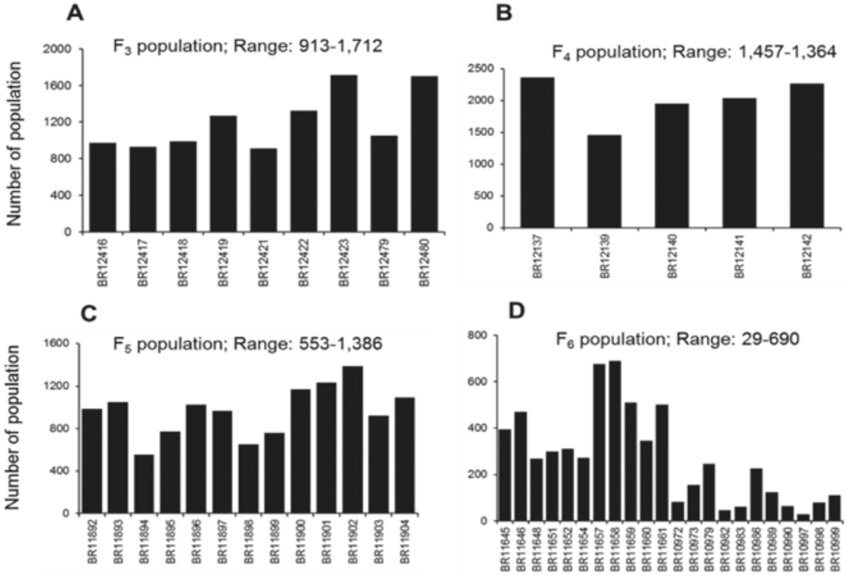

Fig. 3. Histogram showing number of populations produced in F_3 generation (A), F_4 generation (B), F_5 generation (C), and F_6 generation (D) field RGA nursery of CTR during Boro 2018-19.

In 2020, the number of progenies per population varied from 152-492 for F_2, 160-450 for F_4, 122-345 for F_6 under FBR breeding programme (Fig. 4). On the contrary, the number of progenies per population in CTR breeding programme ranged from 108-292 for F_2, 195-436 for F_4, 192-1004 for F_5, and 167-1010 for F_6 (Fig. 5).

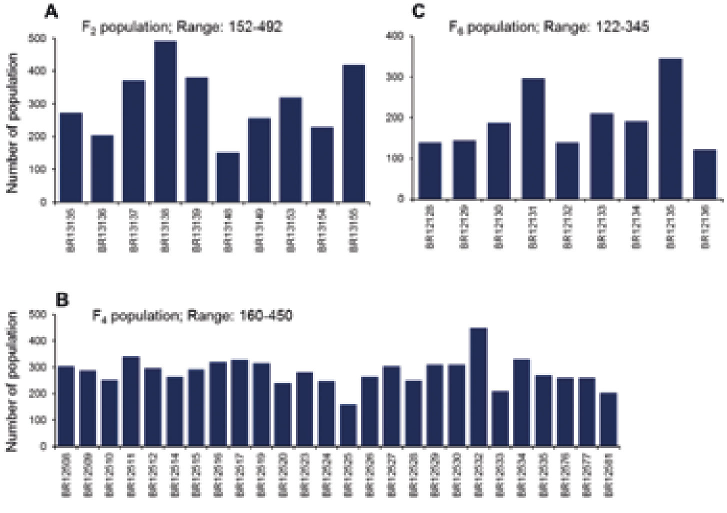

Fig. 4. Histogram showing number of populations produced in F_2 generation (A), F_4 generation (B), F_6 generation (C), field RGA nursery of FBR during Boro 2020.

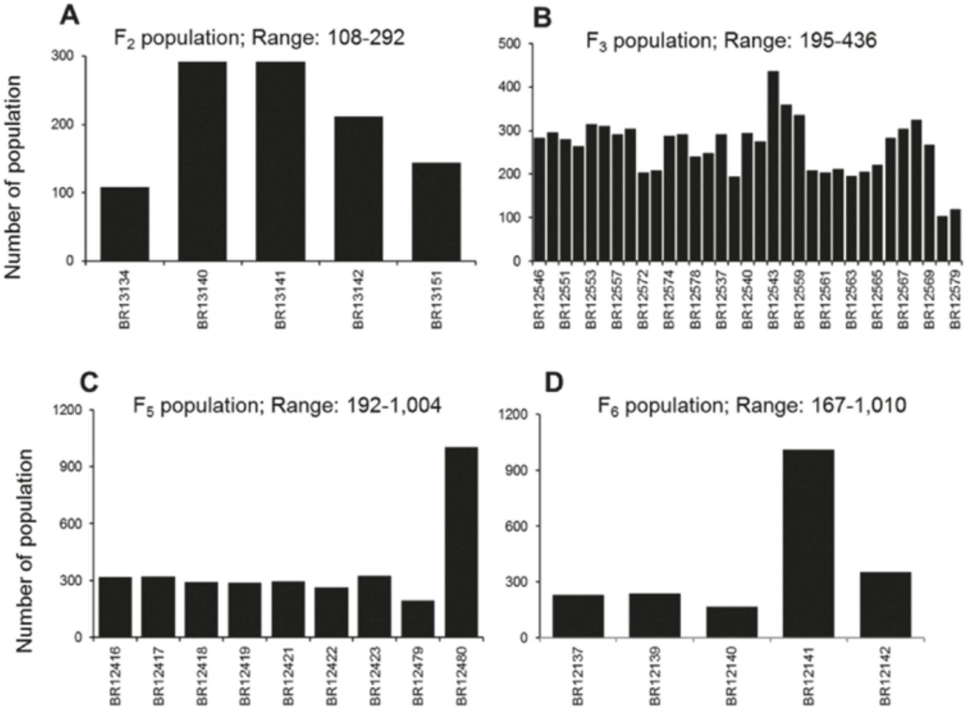

Fig. 5. Histogram showing number of populations produced in F_2 generation (A), F_4 generation (B), F_5 generation (C), and F_6 generation (D) field RGA nursery of CTR during Boro 2020.

Development and selection of fixed lines in Line Stage testing (LST)

LST is a kind of seed amplification stage of F5:6 of F6:7 fixed lines for the next step of field evaluation for yield potential. In this stage, homogeneous lines are isolated. In FBR breeding programme, a total of 8,791 lines were evaluated (Table 2). In each cross, the number of populations ranged from 137-496 lines. Large variation was observed in flowering, plant height and grain size and shape. Final selection was made based on genotyping profiles for BLB, Blast and BPH resistance, and different grain quality traits. Initially progeny selection was performed based on visual phenotypic performance, which followed screening against the presence of favourable alleles for the target key traits.

In FBR breeding programme, 1,959 homogeneous lines were initially identified. The genotypic profiles of these lines showed that majority of the lines (88.5%) had favourable alleles for high amylose specific marker *wx-A, wx-10* and *wx-b*. Out of initially selected lines, 493 and 441 lines had favorable alleles of blast resistant genes *Pb1* and *Pi-ta* (Fig. 6). In addition, 147 lines had favourable allele for BB resistance gene *xa5*. Finally, 406 fixed lines from 23 cross-families were selected for yield evaluation. The number of selected lines per cross ranged from 1 to 86 lines. The selection intensity also varied from 0.21-20.1% across the tested population in LST (Table 2).

Table 2. Selection Summary of RGA derived breeding population in LST under FBR.

Cross name	Developed	Selected	Selection intensity (%)
BR11617	432	9	2.1
BR11619	489	3	0.6
BR11623	493	8	1.6
BR11624	496	3	0.6
BR11626	377	3	0.8
BR11630	475	30	6.3
BR11631	496	21	4.2
BR11634	137	5	3.6
BR11635	231	7	3.0
BR11636	488	23	4.7
BR11637	305	51	16.7
BR11638	427	86	20.1
BR11640	493	61	12.4
BR11641	486	1	0.2
BR11882	376	2	0.5
BR11884	318	5	1.6
BR11885	296	2	0.7
BR11886	365	5	1.4
BR11887	387	21	5.4
BR11888	368	8	2.2
BR11889	333	13	3.9
BR11890	167	16	9.6
BR11891	356	23	6.5
Total	8,791	406	-
Range	137-496	1-86	0.21-20.1

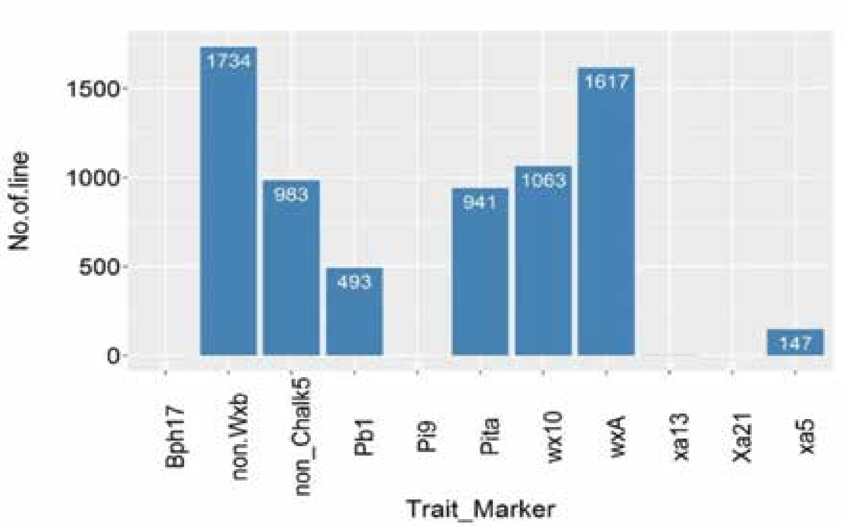

Fig. 6. Frequency of trait makers showing favourable alleles for different key target traits among 1,940 LST lines under FBR breeding programme.

Under CTR, 8,842 RGA derived lines comprising 76 – 567 lines per cross-family were evaluated in LST (Table 3). Large variation was observed in flowering, plant height and grain size and shape. The genotypic profiles showed that among the initially selected 1780 lines, the favourable alleles for Blast resistant genes, *Pita* and *Pb1* were present in 25.2% and 48.0% lines. Moreover, 88.5% lines had favourable alleles for high amylose content, 2.5% for BPH resistance and only 1.0% for BLB resistant gene *xa5* (Fig. 7). Finally, 388 lines were selected based on the presence of favourable alleles for target traits and acceptable visual appearance (Table 3). The number of selected lines ranged between 1 to 49 lines per cross. The selection intensity also varied from 0.60-14.9% across the tested population in LST (Table 3).

Table 3. Selection Summary of RGA derived breeding population in LST under CTR.

Cross name	Developed	Selected	Selection Intensity (%)
BR11645	300	6	2.0
BR11646	433	21	4.8
BR11648	241	36	14.9
BR11651	250	8	3.2
BR11652	267	23	8.6
BR11654	258	8	3.1
BR11657	464	6	1.3
BR11658	567	30	5.3
BR11659	434	49	11.3
BR11660	329	26	7.9
BR11661	471	28	5.9
BR10972	85	5	5.9
BR10973	150	1	0.7
BR10979	272	8	2.9
BR10989	139	3	2.2
BR10998	76	3	3.9
BR10999	100	4	4.0
BR11892	357	5	1.4
BR11894	389	8	2.1
BR11895	354	13	3.7
BR11896	321	38	11.8
BR11897	304	6	2.0
BR11898	254	3	1.2
BR11899	368	5	1.4
BR11900	351	13	3.7
BR11901	347	2	0.6
BR11902	326	9	2.8
BR11903	330	11	3.3
BR11904	305	10	3.3
Total	8,842	388	-
Range	76-567	1-49	0.6 - 14.9

DISCUSSION

Higher number of segregating populations in RGA nursery

BRRI had been using pedigree method for breeding rice since its establishment in 1970. Although, using this breeding method, many varieties of different crops have been developed in many countries; it showed its weakness in identifying superior genotypes aiming to breaking current level of yield ceiling. On the contrary, single seed decent (SSD) method using RGA techniques have shown promise in this regard (Collard *et al.*, 2017). The SSD-RGA not only produces superior progenies but also shorten breeding cycle significantly, which in turn brings a frame-shift change enhancing genetic gain in the breeding population. Very recently, BRRI has adopted this breeding method (Rahman *et al.*, 2019) targeting accelerated genetic gain in different rice breeding programmes including boro rice for favourable and cold prone environments. For a successful breeding programme, existence of wide variations is pre-requisite. RGA method is capable of producing large number of fixed lines from segregating population rendering huge variations between them. In the present study, we also observed wide variation in visual agronomic performances and genotyping profile for key target traits in the LST class fixed lines developed through SSD-RGA techniques.

Firstly, under FBR programme, 62,269 individual progenies from 64 crosses and 12,122 individual progenies from 45 crosses were advanced through RGA in 2019 and 2020, respectively. In contrast, 68,531 progenies from 82 crosses and 15,004 progenies from 52 crosses were advanced under CTR in 2019 and 2020, respectively. These observations clearly indicate that RGA system was capable to produce a large number of segregating progenies (F_2-F_6) in the RGA nurseries. Thus, larger variations can be conserved from elite-by-elite crosses. Use of RGA system for advancing segregating population has been reported in the Philippines, Thailand, India, Bangladesh and Japan (Eunus *et al.*, 1980; Vergara *et al.*, 1982; Das, 2013; Janwan *et al.*, 2013; Manigbas and Lambio, 2015; Maruyama, 1989; Rahman *et al.*, 2019).

In this study, we observed remarkable differences in the distribution of each segregating progenies in both of the breeding programmes. The ranges of progenies were always higher for F_2 population and always lower for F_6 population. For instance, the number of lines per cross in F_2 population ranged between 442-1912 in FBR 2019 whereas it ranged from 33-1060 in F_6 population in the same year. In 2019 under CTR breeding

programme, the F_2 population had 182-1,889 progenies per cross whereas 29-690 for F_6 population under CTR programme. In 2020, there were less variation among number of progenies for each cross under both FBR and CTR programme. In principle, SSD-RGA should produce almost equal number of progenies from F_2 generation to F_6 generation. The reason behind the comparatively a smaller number of progenies of late generations in both the breeding programme might be due to higher rate of mortality or poor recovery rate during RGA process because of inadequate training to the personnel concerned for handling RGA activities and unavailability of optimized RGA protocol. In recent days, situation has been remarkably improved in realizing large number of progenies per cross at each generation. However, only 400 LST lines is good enough for yield evaluation to isolate breeding lines of 2-sd yield advantage over the population mean with combinations of favourable alleles of couple of target traits. With the progression of the breeding programmes in last few years, the breeders in BRRI have enriched their knowledge level in handling RGA population. Now, initial breeding population size in F_2 generation has been lowered to 500 to conserve available variation from the elite-by-elite crosses.

Selection of promising fixed lines in LST

In the population improvement through the RGA system, no selection pressure is imposed on segregating progenies (F_2-F_6). Thus, the LST trial of RGA derived fixed lines has been considered as an important step for seed increase and allowing selection of highly heritable traits. This is the very first step of selection for plant type, disease resistance or other desired traits (Collard *et al.*, 2017, Rahman *et al.*, 2019). At the present study, RGA derived fixed lines were grown in single row consisting of only 10-12 hills. In our experience, this was sufficient to isolate better lines with uniformity in flowering time, grain size and shapes. Under FBR programme, 8,791 lines were transplanted in the field with at least 137 lines for each cross. Considering desirable agronomic characters and presence of favourable alleles for BLB, Blast and grain quality traits, a sum of 406 promising breeding lines were selected from LST trial of FBR. On the other hand, out of 8,842 lines grown with at least 76 lines for each cross under CTR programme, 388 promising breeding lines were selected for further evaluation in replicated field trial. The selection intensity in these breeding programmes varied from 0.21 – 20.1% to 0.60-14.9%. These results indicated that at least 100 RGA derived fixed lines were sufficient to produce possible variations in LST trial. During first selection of RGA derived lines, there were advantages in isolating desired lines based on a panicle-row rather than selecting single plants. We quickly discarded at least 20-30% of the inferior lines with obvious defects by visual observation. Taken together, 794 fixed breeding lines were isolated from 17,633 RGA derived lines under FBR and CTR programme for irrigated Boro rice development. In a separate study of salinity tolerance breeding programme, around 3000-4000 plants per cross was maintained in F_2 to ensure around 400 LST lines per cross. At LST, 2882 elite fixed lines were tested in T. Aman 2018-19. Selection was applied to isolate progeny rows on the basis of our desired traits like homogeneity, growth duration, grain type, disease tolerance, and phenotypic acceptability (Rahman *et al.*, 2019).

Likewise, in the RGA population of IRRI, several 100 lines per population were maintained in fixed line evaluation which is required to increase the likelihood of identifying transgressive segregants (Collard *et al.*, 2017, Snape and Riggs, 1975). Uses of advanced molecular markers have shown great promise in developing high yielding modern varieties with specific trait benefits. Using gene/QTLs specific markers and relevant donor parents in the breeding programme powered by RGA technique could generate future varieties with significant improvement in genetic gain. In LST trial, genotyping was done to figure out the favourable alleles for all desired traits in the breeding population of FBR and CTR. However, SNP-based trait genotyping has showed remarkable efficiency in identifying genetically important lines with multiple stress tolerance including seedling stress salt tolerance, blast resistance and gall midge resistance (Debsharma *et al.*, 2022). In the breeding population designed for irrigated Boro rice, trait marker profiles were considered mainly for amylose, BLB, Blast and BPH resistance. In the LST of FBR, selected 441 lines had blast resistant genes *Pb1* and *Pi-ta*, 147 lines had favourable allele for BB resistance gene *xa5*, while in CTR breeding programme, 88.5% lines had favourable alleles for high amylose content, 2.5% for BPH resistance and only 1.0% for BLB resistant gene *xa5*. These results indicated that the RGA breeding method had better advantage of integrating marker assisted selection than the phenotyping in reduced population sizes after selecting comparably superior lines. Additionally, shortcomings of present trait improvement programme such as lack of favourable allele for *Xa21* (bacterial leaf blight resistance) and *Pi9* (blast resistance) can also be addressed by deploying MAS under forward breeding strategy in the LST class fixed lines.

CONCLUSIONS

The aim of establishing RGA facilities included production of large number of segregating population in each cross, conservation of possible genetic variation developed from elite-by-elite crosses and increase of genetic gain from newly developed RGA derived fixed lines. This study revealed that present RGA system of FBR and CTR programme was able to fulfill the objectives of adopting RGA techniques as a routine activity in the breeding programme for producing large number of segregating progenies (F_2-F_6) for both the programmes. Further, it also confirmed that LST trial of RGA derived fixed lines was able to capture wider variation among fixed lines with wide distribution of multiple traits. Thus, selection was made after possessing high selection pressure in a large set of breeding population. In the future, the rate of genetic gain and trait improvement could be accelerated through the careful implementation of modern breeding techniques including RGA.

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AUTHOR CONTRIBUTIONS

M M E Ahmed, W Afrin, M Y Khan and P S Biswas performed the experiments for rapid generation advance of segregating generations and managed the Lines Stage Testing experiments. M M E Ahmed and W Afrin prepared the leaf samples of selected LST population for trait genotyping through SNP markers. M M E Ahmed, P S Biswas, and MRA Sarker wrote the draft manuscript. P S Biswas and K M Iftekharuddaula revised the final version of the manuscript. All authors read and approved the manuscript.

COMPETING INTERESTS

The authors declare no competing interests.

REFERENCES

Afrin, S, M N Bari, M M Kabir, A B Uddin and S S Haque. 2019. Incidence of major insect pests and natural enemies in three rice growing seasons of Bangladesh. *Bangladesh Rice Journal*, 23 (1): 35-43.

Ahmed, M M E, A Biswas and S Afrin. 2022. Contribution of IR20 and IR64 in Developing Three Bangladeshi Popular Rice Cultivars. *Plant Breeding and Biotechnology*, 10: 81-93. DOI:10.9787/PBB.2022.10.2.81.

Atlin, G N, J E Cairns, and B Das. 2017. Rapid breeding and varietal replacement are critical to adaptation of cropping systems in the developing world to climate change. *Global Food Security*, 12, 31–37. DOI:10.1016/j.gfs.2017.01.008.

BBS (Bangladesh Bureau of Statistics). 2018. Statistical Year Book of Bangladesh.

Ministry of Planning, Government of the People's Republic of Bangladesh, Sher-e Bangla nagar, Bangladesh.

Carandang, F M, S Shanmugasundaram and A L Carpena. 2006. Rapid generation advancement in soybeans using immature seeds. *Philippine Journal of Crop Science*, 31: 53–59.

Cobb, J N, P S Biswas, J D Platten. 2019a. Back to the future: revisiting MAS as a tool for modern plant breeding. *Theoretical and Applied Genetics*, 132 (3): 647-67.

Cobb, J N, R U Juma, P S Biswas, J D Arbelaez, J Rutkoski, G Atlin, T Hagen, M Quinn, E H Ng. 2019b. Enhancing the rate of genetic gain in public-sector plant breeding programs: lessons from the breeder's equation. *Theoretical and Applied Genetics*, 32 (3): 627-45.

Collard, B C Y, J C Beredo, B Lenaerts, R Mendoza, R Santelices, V Lopena, H Verdeprado, C Raghavan, G B Gregorio, V Leigh, M Demont, P S Biswas, K M Iftekharuddaula, M A Rahman, J N Cobb and M R Islam. 2017. Revisiting rice breeding methods – evaluating the use of rapid generation advance (RGA) for routine rice breeding. *Plant Production Science*, 20: 4, 337-352, DOI:10.1080/1343943X.2017.1391705.

Collard, B C Y, G B Gregorio, M J Thomson, M R Islam, G V Vergara, A G Laborte, E Nissila, T Kretzschmar, J N Cobb. 2019. Transforming Rice Breeding: Re-Designing the Irrigated Breeding Pipeline at the International Rice Research Institute (IRRI). *Crop Breeding Genetics and Genomics*. 1-19. DOI: 10.20900/cbgg20190008

Das, S R. 2013. Reflections on >40 years of rice breeding for eastern India. *SABRAO Journal of Breeding and Genetics*, 45: 67–72.

Debsharma, S K, M A Rahman, M R Quddus, H Khatun, R F Disha, P R Roy, S Ahmed, M El-Sharnouby, K M Iftekharuddaulla, S Aloufi, F M Alzuaibr. 2022. SNP Based Trait Characterization Detects Genetically Important and Stable Multiple Stress Tolerance Rice Genotypes in Salt-Stress Environments. *Plants*, 11 (9): 1150.

Fuente, D L, G N Frei, U K and T Lubberstedt. 2013. Accelerating plant breeding. *Trends in Plant Science*, 18: 667–672. DOI:10.1016/j.tplants.2013.09.001

Eunus, M, Ikehashi, H and B S Vergara. 1980. Screening for resistance to bacterial blight of rice under rapid generation advance. International Rice Research Newsletter, 5-7.

Gaur, P M, J Kumar, C L Gowda, S Pande, K H Siddique, T N Khan, T D Warkentin, S K Chaturvedi, A M Than and D Ketema. 2008. Breeding chickpea for early phenology: perspectives, progress and prospects. In: Proceedings of the Fourth International Food Legumes Research Conference. 39-48.

Goulden, C H. 1939. Problems in plant selection. In Proceedings of the Seventh International Genetics Congress. *Cambridge University Press*, pp. 132-133.

Janwan, M, T Sreewongchai and P Sripichitt. 2013. Rice Breeding for high yield by advanced single seed descent method of selection. *Journal of Plant Sciences*, 8: 24–30.

Liu H, P Zwer, H Wang, C Liu, Z Lu, Y Wang, G Yan. 2016. A fast generation cycling system for oat and triticale breeding. *Plant Breeding*, 135 (5): 574-9.

Manigbas, N L and L A F Lambio. 2015. Rapid generation advance in developing recombinant and backcrossed inbred lines for high temperature tolerance in rice (*Oryza sativa* L.). *Rice-Based Biosystems Journal*, 1: 39–48.

Maruyama, K. 1989. Using rapid generation advance with single seed descent in rice breeding. Progress in irrigated rice research: Selected papers and abstracts from the international rice research conference, International Rice Research Institute, Hangzhou.

Ohnishi, T, M Yoshino, H Yamakawa and T Kinoshita. 2011. The biotron breeding system: A rapid and reliable procedure for genetic studies and breeding in rice. *Plant and Cell Physiology*, 52: 1249–1257. DOI: 10.1093/pcp/pcr066.

Rahman, M A, M R Quddus, N Jahan, M A Rahman, M R A Sarker, H Hossain and KM Iftekharuddaula. 2019. Field Rapid Generation Advance: An Effective Technique for Industrial Scale Rice Breeding Program. *The Experiment*, 47 (2): 2659-2670.

Rizal G, S Karki, M Alcasid, F Montecillo, K Acebron, N Larazo, R Garcia, I H Slamet-Loedin and WP Quick. 2014. Shortening the breeding cycle of sorghum, a model crop for research. Crop Science. 54 (2): 520-529.

Saxena, K B, R K Saxena, L T Hickey and R K Varshney. 2019. Can a speed breeding approach accelerate genetic gain in pigeonpea? *Euphytica*, 215 (12): 1-7.

Snape, J W and T J Riggs. 1975. Genetical consequences of single seed descent in the breeding of self-pollinating crops. *Heredity*, 35 (2): 211.

Tanaka, J, T Hayashi and Iwata H. 2016. A practical, rapid generation-advancement system for rice breeding using simplified biotron breeding system. *Breeding Science*, 66: 542–551. DOI: 10.1270/jsbbs.15038.

Vergara, B S, G Patena and F S S Lopez. 1982. Rapid generation advance of rice at the International Rice Research Institute. IRRI research paper series-International Rice Research Institute.

Watson, A, S Ghosh, M J Williams, W S Cuddy, J Simmonds, M D Rey, M H M Asyraf, A Hinchliffe, A Steed, D Reynolds and N M Adamski. 2018. Speed breeding is a powerful tool to accelerate crop research and breeding. *Nature plants*, 4 (1): 23-9.

Bangladesh Rice J.26 (1): 47-57, 2022, doi.org/10.3329/brj.v26i1.65883

Fabrication and Field Performance of Power Weeder for Mechanized Rice Cultivation in Bangladesh

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ABSTRACT

A study was aimed at modifying and manufacturing a power weeder at the local workshop using locally available material and evaluating its performance in the condition of Bangladesh. The Bangladesh Rice Research Institute's Farm Machinery and Post-harvest Technology (FMPHT) Division (BRRI) took the initiative to fabricate a power weeder using locally available materials. In the wetland of the BRRI research field and farmer's field at Jogitola of Gazipur district, the developed power weeder was tested during the Boro season of 2017-2018. The average weeding efficiency of the power weeder was 80.38% and 81.43% at the research and farmers' field respectively. The percent of tiller damage was observed 2.78% and 2.81% respectively. 910 m^2 h^{-1} (0.091 ha h^{-1}) was the average effective field capacity of the power weeder. After five days, the percentage of weeds revived for power weeder was observed at 32.26% and 34.90% at the BRRI research and farmers' fields, respectively. Weed biomass was found 35.43 gm m^{-2} in a farmer's field and 30.88 gm m^{-2} in the BRRI research field, Gazipur. This machine can be run by one man/woman easily. The weight of the complete weeder is 18.3 kg. The benefit-cost ratio of the weeder is 1.85. Farmers can use this weeder in wetland conditions. In the line transplanted wetland field conditions, the power weeder was found suitable for controlling weeds with minimum standing water.

Key words: Power weeder, fabrication, plant damage, weeding efficiency, field capacity, field efficiency.

INTRODUCTION

Weeds compete with the crop for water, light, and plant nutrients rather than harboring insects, and adversely affect the microclimate around the plant. Weeds extract 30-40 percent of the applied nutrients in the absence of an efficient control measure, resulting in a substantial reduction in yield. Mechanical weeding is preferred because manual weeding is time-consuming, tedious, and costly. Mechanical weeding is done either by a power-operated weeder or a manually-operated weeder. Manually operated weeders have found acceptability due to their low cost but involve drudgery. Weed control demands a lot of human labour, sometimes several weeding is required to keep the crop weed-free. Chinnusamy *et al.*, 2000 stated that it was necessary to maintain a weed-free cycle for up to 45 days after transplantation to increase medium-term rice yields. About 30-60 days after the sowing cycle in rain-fed lowland rice was considered as a crucial period for crop weed competition to avoid losses of grain yield (Moorthy and Saha, 2005). Singh *et al.*, 2002 found that retaining weed-free status until maturity resulted in

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substantially higher grain yield due to more panicles per m2 and lower weed density and dry weight.

Weeds have a significant negative impact on crop production and are responsible for marked losses in crop yields and faster root and shoot growth abilities than the crop (Mamun *et al.*, 1993). Manual weeding requires a large labour force and accounts for around 25 percent (900-1200 man-hours/hectare) of the total labour requirement (Nag and Dutt, 1979). Depending on the crop and location, the reduction in yield due to weeds alone is estimated to be 16-42 percent and involves one-third of the cultivation expense (Rangaswamy *et al.*, 1993). In paddy production, weeds are the key restriction and a direct determinant of crop yield reduction. Weeds reduce yields from 40 percent to 65 percent, and the most significant problem facing farmers is their eradication.

In Bangladesh, the yield loss due to weed competition in Aman rice is 40%. (BRRI, 1991). Weeds in Bangladesh are manually managed by pulling or using simple tools such as niranee, Japanese rice weeder, BRRI weeder, etc. Generally, according to the nature of the weeds and the severity of infestation, two to three hands of weeding are performed for growing transplanted rice crops. These strategies, however, are laborious, less convenient, time-consuming, and costly as well. The cost of mechanical weeding is almost 30 percent to 50 percent less than hand weeding, Atajuddin, 2004 estimated. It can be eliminated by hand weeding, by chemical means, by the use of herbicides, or by mechanical weeding. Hand weeding is the most effective form of weeding, but due to greater time consumption coupled with labour-intensive activity and expense, it is not well suited. The chemical method shows promising results in the eradication of weeds, but due to its poor impact on humans and the climate, it is limited. As a result of improved soil aeration, root length, and better tiller efficiency, mechanical weeding encourages plant growth. A conventional hand-aided weeding instrument may do this; mechanical weeders and power weeders are manually operated.

A power weeder was developed, evaluated and performance was compared with traditional weeding with a manually operated dry land weeder hoe (Rangasamy *et al.*, 1993). The weeder's field capacity was 0.04 ha/h with a 93% weeding efficiency. The operating cost of the power weeder was 250/ha compared to 490/ha for the dry land weeder and 720 for manual weeding with a hoe. The time and cost savings were 93% and 65%, respectively. An engine-operated rotary weeder with a 'L' shaped cutting blade device for wetland paddy has been developed and developed as a recommendation for weed control (Victor *et al.*, 2003). The different methods used in the process are manual, biological, chemical, and mechanical weeding. Each approach has its advantages and drawbacks, whereas the advantages of mechanical weeding are commonly used. Chemical weeding can cause environmental impacts, although no pollution is caused by the mechanical process. The demand for good quality food on the market is very strong, now people are willing to pay some extra amount a day if the quality is guaranteed. Farmers have to build processes and mechanisms for the development of quality crops and end-user goods to meet consumer demand (Patil *et al.*, 2018).

Since the time available for weeding is minimal, improved mechanical weeders should be used at a minimum cost to complete the weeding process in due time. Due to concern about environmental degradation due to herbicide usage and rising demand for organic food, there is an increasing interest in the use of mechanical weeders. To ensure food security and pollution-free climatic conditions, the agricultural sector needs non-chemical methods of weed control. Weeds can be managed by mechanical weeders in a manner that meets user and environmental and pollution-free requirements. Mechanical methods of weed control ensure safety against soil and water contamination as well. The majority of Bangladeshi farmers in the rice field manage weeds by hand weeding. In addition to pulling the weed between the crop rows, mechanical weed control often makes the soil surface lose, ensuring better aeration of the soil and water intake capacity. Under the 'BRRI-Project "Development of research capacity of the Bangladesh Rice Research Institute" a Korean power weeder was collected, which was suitable for a mechanical transplanted field of 30 cm line spacing. It was changed to fit the 18, 20, and 22 cm line spacings used in Bangladesh (Hossen *et al.*, 2015). The modification was done in a rotary drum. The only width of the rotary drum was reduced and other parts of the weeder were the same as the Korean weeder. In a single-pass operation and operated by a petrol engine, the power weeder was fitted with three rotors to weed out three rows. All parts and engine of the Korean weeder were not available in Bangladesh. Under this circumstance, an attempt was taken to fabricate all parts (Fig. 1) of the power weeder by using locally available materials. Considering the above points, the experiment of the fabrication of a power weeder using locally available materials and field performance of the fabricated weeder for mechanized rice cultivation in Bangladesh condition has been undertaken.

MATERIALS AND METHOD

Development of power weeder

FMPHT Division of BRRI has been updated for 18, 20, and 22 cm line spacing under the KOICA-BRRI project (2012) where the Korean power weeder is suited for the rice field line spacing of 30 cm. At that time only the weeder was modified for 18-22 cm spacing. After that, again FMPHT Division, BRRI took the initiative to develop and fabricate this weeder using locally available raw materials. For that intention, the FMPHT Division fabricated a Korean model power weeder under a public-private partnership (PPP) at the Alam engineering workshop in Dhaka. In this workshop, the weeder was manufactured as per design. The original specification of the power weeder was reviewed during design. All parts of the weeder were fabricated under this workshop using locally available materials. GI pipe, GI board, MS sheet, MS flat bar, MS shaft, etc workshop materials were used in the workshop to manufacture the weeder. During the Boro 2018 season at the BRRI research field and farmer's field at Jogitola, Gazipur, a developed weeder was tested.

Description of the fabricated power weeder

In a single-pass operation, the fabricated power weeder was fitted with three rotors to weed out three rows and driven by a petrol engine. Table 1 presents the specifications of the developed power weeder. Major components of the newly fabricated power weeder were the engine, worm gear, spline shaft, rotor, spike, and frame.

A small petrol engine is used to power it (1.47 kW @ 7000 rpm) which was used as the main power source. The power from the engine was transmitted by a coupling mechanism. This power was transmitted to the spline shaft, which is engaged and disengaged with the rpm rate. Engage and disengage between the engine main shaft and propeller shaft is done by clutch plate type coupling mechanism. The high rpm of the engine was reduced by the worm gear to get the desired rotor rpm. In a single pass, it covers the 60 cm width of the paddy field. For the weeding, single and triple spike plates were used. The rotor's weeding spike was made of MS sheet, and the weeder's rotor was made of aluminum sheet. One man or woman can comfortably operate this machine. The weight of the weeder is 18.3 kg in total.

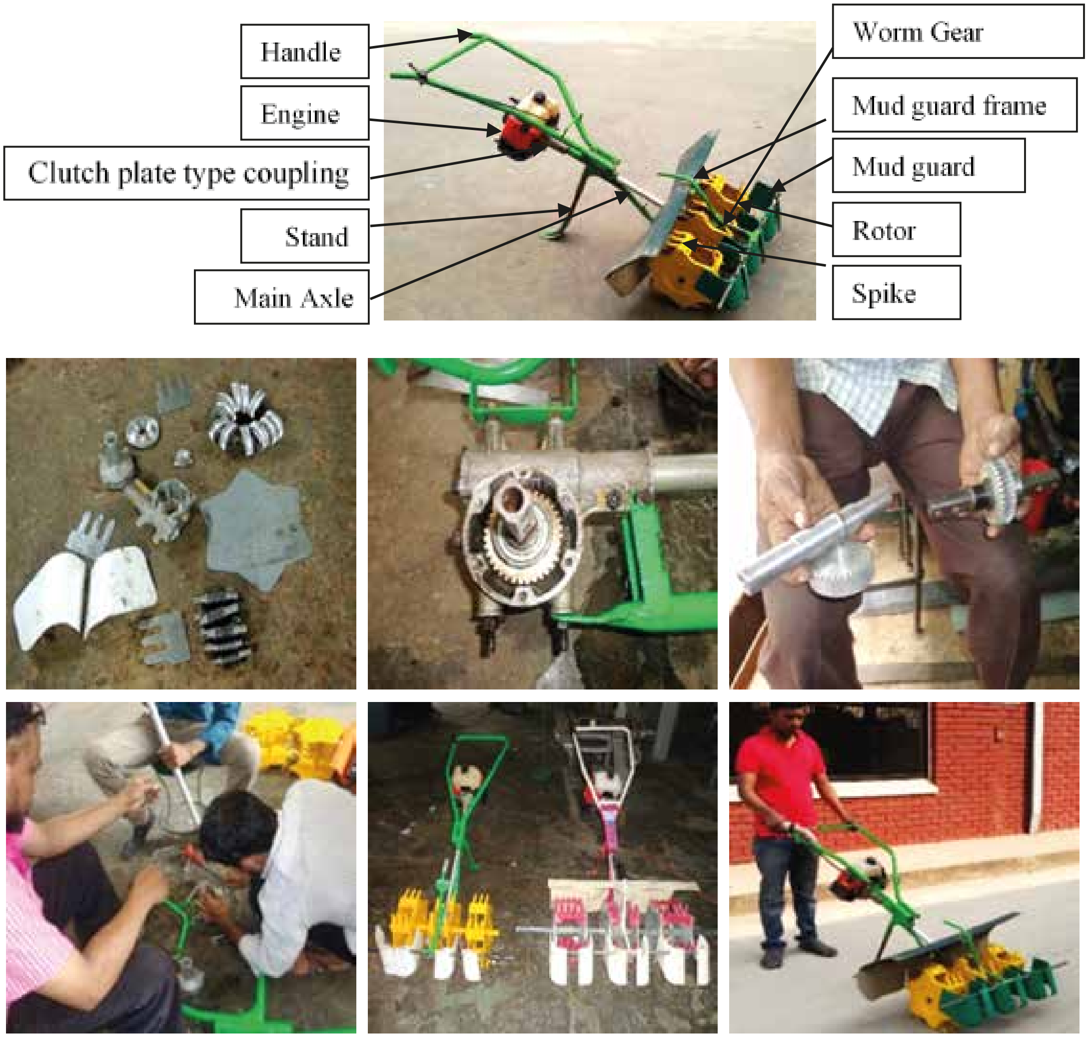

Fig. 1. Complete view of the fabricated power weeder with different parts of the weeder.

During the study, the following data were reported and measured.

- Speed for walking, m/sec.
- Weeding time, min.
- Time spent on turning, min
- Fuel consumption, l/hr.
- Actual field capacity, m^2/hr.
- Theoretical field capacity, m^2/hr.
- Field efficiency of the weeder, %
- Number of weeds before weeding
- Number of weeds after weeding 0, 5, and 10 days
- Number of tiller before weeding
- Number of tillers after weeding
- Weeding efficiency, %
- Weed biomass
- Number of tillers after weeding

OPERATIONAL PROCEDURE

To calculate the theoretical field capacity of the weeder, walking speed was recorded without any loss. Total field operation time was reported to calculate the weeder's actual field capacity with turning loss, operator loss, and loss during field operation for system adjustment and troubleshooting losses. Before and after a field operation, the number of weeds and tiller numbers were recorded from the pre-selected 1m^2 areas. Weeds were also collected from an area of 1 m^2 to assess the weed biomass before weeding. Collected weeds were dried in an oven at 75°C for 48 hours. To calculate weeding capacity, weeding efficiency, and the amount of tiller/hill injured, the following formula was used.

FIELD CAPACITY

The actual field capacity of the fabricated power weeder was measured during operation in the study locations. To measure the actual field capacity of the weeder, the machine operating period included the time needed during the weeder's turning, the operator's time, adjustment time, re-starting time, etc. It is the proportion of the machine's real average field coverage rate to the total time during operation (Hunt, D. 1995). Therefore,

$$C = \frac{A}{T} \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots \text{(1)}$$

Where,
C = Actual field capacity in ha/hr.
A = Area of weeding in hector
T = Time of weeding in hr.

WEEDING EFFICIENCY

The average number of weeds present per square meter area before weeding should be determined. Similarly, the number of weeds left out per square meter can be counted five days after the weeding test is completed. The difference between the two will give the number of weeds eliminated and the efficiency of the weeder can be computed using the following equations (Remensan *et al.*, 2007).

$$\textbf{Weeding efficiency} = \frac{\textit{Number of weedes eliminated per } m^2}{\textit{Total Number of weeds present per } m^2} x\ 100$$

$$WE = \frac{W1 - W2}{W1} \text{ X } 100 \ldots\ldots\ldots\ldots\ldots\ldots\ldots \text{(2)}$$

Where,
WE = Efficiency of weeding in percentage
W1 = Population of weeds before the operation
W2 = Population of weeds after the operation

Damaged tiller rate

The percentage of rice tiller breakage was determined using the following equation:

$$DTR = \frac{T1 - T2}{T1} \text{ X } 100 \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots \text{(3)}$$

Where, DTR=Damage of tiller in percentage
T1 = Tiller number before weeding
T2 = Tiller number after weeding

Table 1. Specification of fabricated power weeder.

Item	BRRI fabricated Power Weeder
Machine type	Walking
Motion type	Forward
Engine type	Petrol engine
Start mode	Exclusive cartridge starting, recoil type
Power Transmission system	The centrifugal clutch → Worm gear (Reduction ratio 1/35)
Weight, kg	18.3
Dimension (L×W×H), cm	140×60×30
Number of rotors	3
Rotor diameter, cm	29
Rotor width, cm	10
Single spike plate number in the middle rotor	12
The number of the middle rotor's double spike plate	0
Triple spike plate number in the lateral rotor	12
The number of a five-spiked plate in the side rotor	0
The cover plate number	6
The dimension of the handle to be carried, cm	40
Stand height, Cm	53

Engine:

Parameter		Observation/Declaration
Engine		
Type	:	Air-cooled, 2-stroke, single-cylinder, Spark Ignition engine
Make	:	Rabbit
Model	:	EC04EA-2
Power, (kW) (apa)	:	1.47 kW @ 7000 rpm

FIELD CONDITION

Table 2. Condition of the field during field operation.

Parameter/Item	BRRI, Research field	Jogitola, Gazipur
Type of Soil	Clay loam Soil	Clay Soil
Depth of standing water (cm)	3-5	4-6
Type of predominant weed	*Scirpus maritimus*	*Scirpus maritimus*
Size of weeds (cm)	15-18	17-21
Stage of maturity of crop, days	20	25
Row spacing of crop, cm	20	20
Plant height (cm)	22-25	28-32

RESULTS AND DISCUSSION

Weeding efficiency

The efficiency of weeding was determined based on the density of the weeds before and after weeding for power weeders. Efficiency in weeding was found 80.38 and 81.43 percent for PW in the BRRI research field and the farmer's field at Jogitola, Gazipur respectively (Fig. 2).

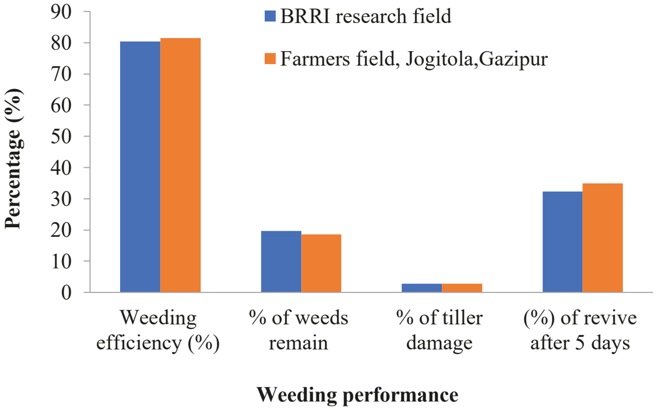

Fig. 2. Weeding performance at BRRI research field and farmers field at Jogitola, Gazipur.

Tiller damage

Tiller damage was observed for PW 2.75 percent at the BRRI research field whereas it was 2.81 percent in the farmer's field at Jogitola, Gazipur. Percent of tiller damage for power weeder was observed higher in farmer's fields because plant height was more and the number of tillers was also more (Fig. 2). Average tiller damage was found 2.78 percent.

Weeds revive

After five days, the percentage of weeds revived for power weeder was observed at 32.26 and 34.90 % at the BRRI research and farmers' fields, respectively. From fig. 2, it was observed that the percentage of weeds revival after five days was high at the farmer's field because the BRRI power weeder could not uproot the weeds properly due to more age and height of weeds at the field.

Field Capacity

The field capacity of the developed power weeder during field activity was calculated in two locations in the Gazipur district. The power weeder's theoretical and actual field capacity were measured during operation to calculate the field efficiency. Theoretical field capacity varied with the forward speed of the operation of the weeder, while actual field capacity varied with the condition of the soil, soil softness, density of weeds, forward speed, loss of turning time, etc. The actual field capacity of the power weeder was found 905 $m^2 h^{-1}$ in the BRRI research field whereas it was 915 $m^2 h^{-1}$ in the farmer's field, Gazipur (Fig. 3). The reason for having higher field capacity in the farmer's field compared to BRRI research field is that in the farmer's field soil was clay loam and that of BRRI research field was heavy clay. It reveals that the machine operation in lighter soil is easier than that of heavy clay soil. The average field capacity was found 910 $m^2 h^{-1}$(0.091 ha h^{-1}). Whereas Hossen et. al (2015) reported that the field capacity of the power weeder was 935 $m^2 h^{-1}$ and the field capacity of the power weeder was 0.08 ha h^1 obtained by Alizadeh, 2011.

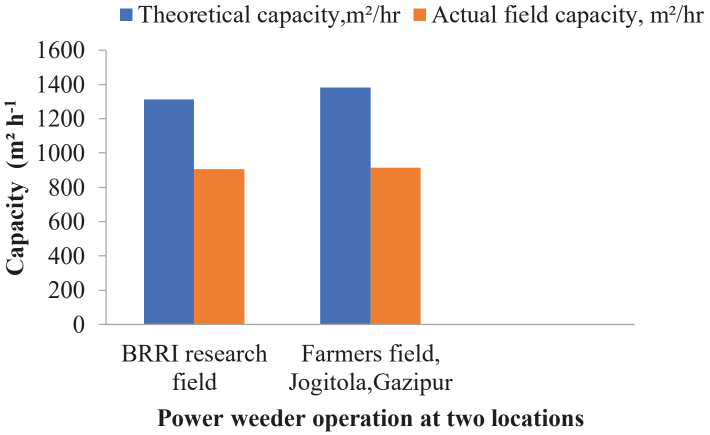

Fig. 3. Field capacity of power weeder.

Field efficiency

The field efficiency of the technologies varied with the variation of total turning time losses. 68.7 % field efficiency was found for PW in the BRRI research field whereas it was observed 70.52 % in the farmer's field respectively (Fig. 4). Average field efficiency was found at 69.61%. Whereas Hossen *et al.* (2015) reported that the field efficiency of the power weeder was 71.37% and 72.36% at Gazipur and Kushtia respectively. The weeder's weeding efficiency (WE) depended on weed severity, soil moisture, weeding regime, operator conditions, and soil conditions. Field efficiency was found lower for PW due to more turning loss and other time loss.

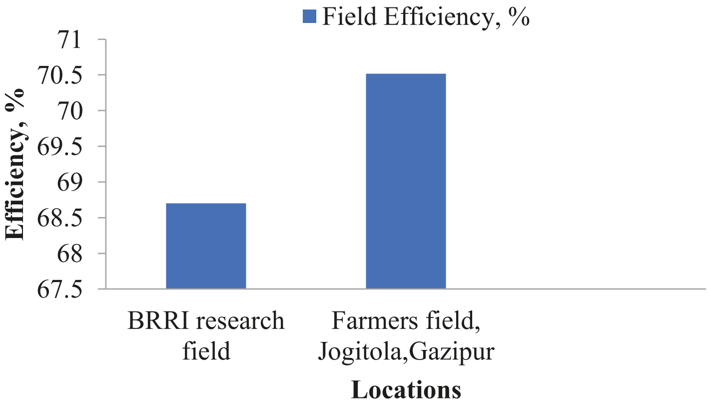

Fig. 4. Field efficiency of power weeder.

Weeds biomass

To observe the real condition of the weeds in the paddy fields, weed biomass was assessed. The number, type, and maturity of the weeds varied in terms of weed biomass. Weed biomass was observed 35.43 gm m^{-2} in a farmer's field and 30.88 gm m^{-2} in the BRRI research field, Gazipur (Fig. 5).

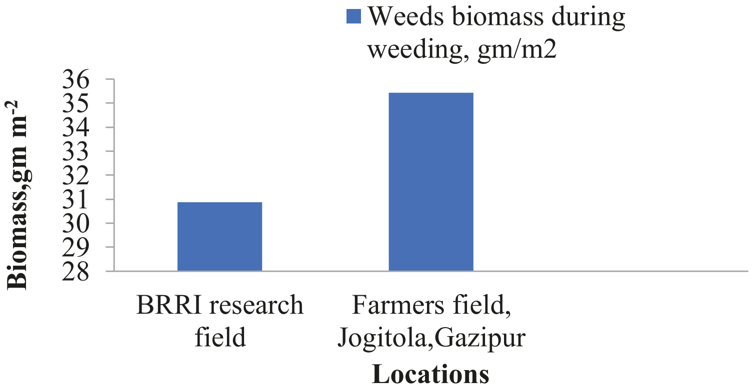

Fig. 5. Weed biomass at different fields.

Financial analysis of BRRI power weeder operations

Weeding operation is done seasonally for rice crop. The total cost of power weeder operations at the farm level included variable costs and fixed costs. Depreciation of the machine was calculated by the straight-line method and taken as a fixed cost. The fixed cost for power weeder operation was estimated 24.96 Tk ha^{-1}. The variable cost for power weeder operation was also estimated 3257 Tk ha^{-1}. Based on field data, the power weeder's operating cost and effective field capacity were found 3531 Tk ha ha^{-1} and 0.091 hectares per hour respectively. The rent-out charge was 6531.47 Tk ha^{-1}. So, the benefit-cost ratio for the power weeder was found 1.85. The results noticed that investment in power weeder was profitable for an entrepreneur. The major cost and return items of the BRRI power weeder are as follows (Table 3).

Table 3. Estimated major cost and return items of power weeder machine operation.

Parameter	Fabricated power weeder
Purchase price (Tk)	30,000
Depreciation, Tk yr^{-1}.	5400
Annual use in the area, ha yr^{-1}.	43.68
The economic life of the machine, yr.	5
Effective field capacity, ha h^{1-}.	0.091
Total fixed cost, Tk ha^{-1}	24.96
Total variable cost, Tk ha^{-1}	3257
Total operating cost, Tk ha^{-1}	3531
Payment for replacement, Tk yr^{-1}.	4422.53
Rent out charge, Tk ha^{-1}	6531.47
Revenue, Tk yr^{-1}.	285294.53
Benefit-cost ratio, BCR	1.85

Note: Average workday =8 hr. at 0.091 ha per hr. or approximately 22.48 decimal per hr.; Price of fuel=90 Tk/liter; labour /operator charge= 500 Tk/day

CONCLUSION

The power weeder was fabricated by using locally available materials and conducted its performance tests in two different locations. A small petrol engine operates the fabricated weeder. This covers 60 cm of paddy field width in a single pass. In the line-transplanted field, the power weeder was found suitable for controlling weeds. The average wedding efficiency of the power weeder was good but the percentage of tiller damage was high. The weeder can uproot, cut, and bury the weeds in a triple row at a time. Moreover, farmers can use this weeder in their fields to get more comfortability in weeding and mulching.

RECOMMENDATIONS

Based on the evaluation result the following recommendations were made:

- The power weeder is recommended for small and medium-scale farmers.
- The weeder can be used in a field with uniform intra row spacing provided the plant is of uniform height.
- The weeder is recommended for weeding in a field with uniform inter and intra-row spacing.
- The weeder should be operated by a physically strong man.
- The rotating shaft of the weeder's blade should be checked regularly to prevent clogging during operation.
- The weeder is recommended for use in tilled farm land.
- The developed power weeder needs to be evaluated in other different soil conditions of the country to find an outfield problem.

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REFERENCES

Alizadeh, M R 2011. Field performance evaluation of mechanical weeders in the paddy field. *Scientific Research and Essays*, 6 (25): 5427-5434.

Atajuddin, F 2004. Mechanizing Indian Agriculture. *Kisan World*, 31, No 6, p 46.

BRRI (Bangladesh Rice Research Institute). 1991. Annual Report for 1991. Bangladesh Rice Res. Inst., Joydebpur, Gazipur, Bangladesh. pp. 45-46.

Chinnusamy, C, Kandasamy, O S, Sathyamoorthy, K And C N Chandrasekar. 2000. Critical period of crop weed competition in lowland rice ecosystems. *In:* Proceedings of State level seminar on Integrated Weed Management in the new millennium, Ratnagiri, Maharastra. 27-28 Feb. 2000.

Hossen, M A, M A Alam , S Paul and M A Hassain. 2015. Modification and evaluation of a power weeder for Bangladesh condition. *Eco-friendly Agril. J.* 8(03): 37-46.

Hunt, D. 1995. Farm Power and Machinery Management, Cost determination, 9th edition, Iowa State University Press, USA.

Mamun, A A, S M R Karim, A K M Hoque, M Begum, M M Kamal and M I Uddin. 1993. Weed survey in wheat, lentil, and mustard crops under Old Brahmaputra Floodplain and Young Brahmaputra and Jamuna Flood Plain Agro-ecological zones. Bangladesh Agril. Univ. Res. Prog. 7: 160-172

Moorthy, B T S and S Saha. 2005. Studies on crop weed competition in rainfed direct-seeded lowland rice (*Oryza sativa* L.). *Indian Journal of Weed Science*, 40(3-4): 112-116.

Nag, P K and P Dutt. 1979. Effective of some simple agricultural weeders with reference to physiological responses, *Journal of Human Ergonomics*, 13-21.

Patil, D P, B B Mujawar, S D Savekar, S J Patil, A R Chougale, S B Porlekar. 2018. Mechanical Power Weeder- Design and Development. Novateur Publications Journal lNX- A Multidisciplinary Peer Reviewed Journal ISSN No: 2581-4230 VOLUME 4, ISSUE 4.

Remensan, R, M S Roopesh, N Remya and P S preman. 2007. "WetLand Paddy Weeding- A Comprehensive Comparative Study from South India". *Agricultural Engineering International: the CIGR E-journal*. Manuscript PM 07011. Vol. IX. December

Rangasamy, K, Balasubramanium M and Swaminathan K R. 1993. Evaluation of power weeder performance, *Agricultural Mechanisation in Asia, Africa and Latin America*, Vol. 24, No.4: 16-18

Singh, R K, S N Sharma, R Singh and M D Pandey. 2002. Efficacy of method of planting and weed control measures on nutrient removal of rice (*Oryza sativa* L.) and associated weeds. *Crop Research*, 24 (3): 425-429.

Victor, V M and A Verma. 2003. Design and development of power-operated rotary weeder for wetland paddy. *AMA.*; 34 (4): 27-29.

Bangladesh Rice J.26 (1): 59-68, 2022, doi.org/10.3329/brj.v26i1.66595

Effect of Polythene Covering on Seedling Quality and Its Carryover Effect on Field Duration and Grain Yield of Rice

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ABSTRACT

Boro rice cultivation is often limit due to lack of a farmer's friendly technique for raising quality seedling in irrigated ecosystem. Experiment was conducted in Boro 2019-20 at BRRI RS, Rangpur to compare different polythene covering treatment on raising quality seedling of BRRI dhan88 and BRRI dhan89 and to measure its carryover effect on growth duration and grain yield of rice. The treatments were as T_1: Day polythene cover (from10:00 am to Sunset), T_2: Night polythene cover (Sunset to Sunrise), T_3: Day-night polythene cover but round shape opening (30 cm diameter) at both sides and T_4: No polythene cover (control). Seedbed was covered by transparent polythene from seeding to 30 days after seeding (DAS). Seedling strength was higher on 01 December seeding than 15 December in both the tested varieties. Day cover always had the lowest seedling strength in both the varieties. Day-night polythene cover treatment (T_3) produced significantly tallest seedling than the other polythene covering treatments for both the planting dates. Seedling mortality was higher in 30 January planting than 15 January planting for both the tested varieties due to prevailing low temperature (below 10°C for eight days). In 15 January planting, BRRI dhan88 and BRRI dhan89 produced higher number of tillers with day cover and night cover treatment, respectively. In 30 January planting, BRRI dhan88 produced higher tiller with day-night and control treatment but day cover had the lowest. Tiller production rate was sharply increased from 35–45 DAT and then decreased. Up to 45 DAT, it was statistically similar in both the varietiers with all treatments. Although, tiller number was higher in T_1, T_2 and T_4 than T_3 but productive tiller (%) was satistically similar among the treatments for both the varieties and planting dates. There was no significant difference in grain yield among the treatments for planting dates and varieties. Day-night polythene cover treatment (T_3) reduces growth duration by 2-3 days over other treatments. This treatment (T_3) is farmer's friendly for raising quality seedling in cold prone areas of Bangladesh.

INTRODUCTION

Temperature is one of the major factors for seedling growth especially in northern part of Bangladesh during Boro season. Northern part is relatively cooler and farmers' normally transplanted Boro rice from last week of January to February with aged seedling. Those who seeding at 1st week of December, seedlings are not affected by cold but who seeding from 2nd week of December, seedlings are affected by cold and became unable to transplant Boro rice timely. Low temperature in vegetative stage can cause slow growth and reduce seedling vigour (Ali *et al.*, 2006), lower number of seedlings (i.e. poor establishment), reduce tillering (Shimono *et al.*, 2002) increase plant mortality (Farrell *et al.*, 2006, Baruah *et al.*, 2009, Fujino *et al.*, 2004), increase the growth period (Alvarado and Hernaiz 2007). The optimum temperature for rice growth and development is 25-30°C. When temperature below as 10-12°C, often cause injury to vegetative organs, creating leaf chlorosis or

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damping-off in rice plants (Tajma, 1995). Quality seedling influences not only the speed of initial growth but also rooting especially under low temperature in transplanting time (Nishiyama, 1995). During cold spell, drain out the water at morning and again irrigate the seedbed, to remove dew on upper part of seedling and seedbed cover with transparent polythene from 10:00 am to sunset may reduce the cold injury in seedlings. But all are not farmers' friendly and there is also some controversy in seedbed covering by transparent polythene sheet at day or night time. So, it needs to develop a farmers' friendly seedling protecting technique during Boro season of Bangladesh.

MATERIALS AND METHODS

A field experiment was conducted at Bangladesh Rice Research Institute, Regional Station, Rangpur experimental farm during winter and irrigated ecosystem in 2019-20. Sprouted seed (80g m^{-2}) were seeded in the puddled seedbed on 1 and 15 December 2019. Seedbed size was 1.0 m × 3.0 m. Seedling age of normal seedbed (control) was also 45 days. No fertilizer was use in seedbed. Experiment was laid out in a factorial randomized complete block design (RCBD) with three replications using plot size of 3.0 m × 3.0 m. The treatments were as T_1: Day polythene cover (from 10:00 am to Sunset), T_2: Night polythene cover (Sunset to Sunrise), T_3: Day-night polythene cover but round shape opening (30 cm diameter) at both sides and T_4: No polythene cover (control). Seedbed was covered by transparent polythene from seeding to 30 DAS (days after seeding). Seedlings were hardened from 30 DAS to 45 DAS. Forty-five-day-old seedlings were transplanted at a spacing of 20 cm × 20 cm with single seedling per hill at 15 and 30 January 2020. At the time of transplanting, fertilizer was used as 120 kg N, 18 kg P and 75 kg K, 25 kg S and 3.6 kg Zn per hectare. Fertilizer N was splitted apply at 20, 35 Days After Transplanting (DAT) and before panicle initiation, respectively. Fertilizer P, K, S and Zn was applied at basal. Weeds, insects and diseases were controlled as when required to avoid yield loss.

Measurements and methods

Seedling strength Ten seedlings were randomly uprooted from each seedbed at 45 DAT in both the planting dates. Then the seedling height (cm) was measured from the base to tip of the seedling. Root of the seedlings were cut and removed from the base and oven dried at 70°C for 72 hrs and dry matter (mg) was weighted. Seedling strength was calculated from the following formula:

Seedling strength (mg/cm) = Dry weight of seedling (mg)/height (cm) of seedling

Seedling mortality (%): (Number of dead seedling in a plot/total number of seedling in a plot)*100. It was taken at 25 DAT in both the plantings.

Tillering pattern: It was started from 25 DAT and continues up to 55-65 DAT with 10 days interval. It was counted 2 × 2 hills from three spot in each plot and expressed as tiller number m^{-2}.

Productive tiller rate (PTR) was calculated as: (Number of panicles m^{-2} /number of maximum tillers m^{-2}) × 100.

Growing degree days (GDD) was calculated following Rajput (1980).

GDD = Σ [(Tmax + Tmin)/2-Tb]. Here, (Tb = Base temperature = 10^{o}C). Where, Tmax = Maximum temperature, Tmin = Minimum temperature.

Plant height: It was measured from the base of the plant to tip of the panicle at harvest.

The crop reached maturity when 90% of the spikelets turned from green to yellow.

Data analysis

Statistical analyses were performed using Statistix 8, Analytical software, Tallahassee, FL, USA. Means of cultivation methods were compared according to the least significant difference test (LSD) at the 0.05 probability level. Figures were performed using MS Excel 2003.

RESULTS AND DISCUSSIONS

Ambient and inner air temperature of different polythene cover treatment

Cold didn’t affect the seedling of 1 December seeding. So, outer and inner temperature was recorded from 15 December seeded plot.

Day vs day-night cover

Inner air temperature of day cover (T_1) was always higher than day-night cover (T_3) at 11:00am. It was 3.4°C and 2.1°C higher than ambient air temperature and day-night cover, respectively. Day-night cover had 1.3°C higher than ambient air temperature. Similarly, inner air temperaturewas 1.2°C and 0.6°Chigher than ambient air temperatureand day-night cover, respectively, at 5:00 pm. Day-night cover had 0.6°C higher than ambient air temperature (Table 1).

Table 1. Difference of ambient air and inner temperature of seedbed by polythene cover, Rangpur, Boro 2019-20.

Day	Temperature (°C)					
	11:00 am			5:00 pm		
	Outside polythene cover	Inside polythene cover		Outside polythene cover	Inside polythene cover	
		Day cover	Day-night cover		Day cover	Day-night cover
15 Dec 2019	24.0	26.0	26.0	22.0	23.0	23.0
16 Dec 2019	20.0	22.0	20.0	18.0	20.0	18.0
17 Dec 2019	21.0	24.0	21.0	18.0	18.0	18.0
18 Dec 2019	15.5	16.5	16.3	16.5	17.0	16.8
19 Dec 2019	14.8	15.5	15.0	15.0	16.0	15.8
20 Dec 2019	14.5	15.0	14.5	14.5	15.0	14.8
21 Dec 2019	14.0	14.5	14.2	14.5	15.0	14.7
22 Dec 2019	16.0	16.5	16.2	16.0	17.0	16.8
23 Dec 2019	19.0	20.0	19.8	18.8	20.0	19.5
24 Dec 2019	16.4	19.5	19.0	19.5	21.0	20.0
25 Dec 2019	21.3	26.0	22.0	18.4	19.4	18.8
26 Dec 2019	14.5	17.0	17.0	14.0	16.0	14.3
27 Dec 2019	19.0	23.0	21.5	17.0	17.8	17.5
28 Dec 2019	14.5	17.5	17.0	18.3	18.8	18.5
29 Dec 2019	22.0	27.0	23.0	18.0	19.5	18.3
30 Dec 2019	24.0	25.0	24.5	22.0	23.0	21.2
31 Dec 2019	25.0	30.0	26.0	20.5	23.0	22.5
01 Jan 2020	22.0	26.0	22.5	20.0	22.0	21.0
02 Jan 2020	21.5	27.0	23.0	18.5	19.5	19.0
03 Jan 2020	17.0	19.90	19.6	15.0	16.0	16.0
04 Jan 2020	21.0	27.0	22.0	21.0	22.0	21.5
05 Jan 2020	19.0	23.0	21.0	18.4	20.5	19.8
06 Jan 2020	15.5	20.0	17.8	13.5	15.5	15.0
07 Jan 2020	19.0	24.0	20.5	19.6	20.5	19.8
08 Jan 2020	21.8	20.0	22.2	20.4	20.8	20.6
09 Jan 2020	24.0	29.0	24.0	22.0	23.0	22.5
10 Jan 2020	18.0	21.5	20.5	17.0	18.0	17.5
11 Jan 2020	16.0	22.0	18.5	20.0	22.0	21.0
12 Jan 2020	16.0	20.0	20.0	17.0	19.0	20.0
13 Jan 2020	23.2	29.0	23.5	19.0	20.5	19.5
14 Jan 2020	24.5	31.0	25.0	20.0	22.0	21.0
15 Jan 2020	28.0	31.5	29.5	20.0	21.0	20.8
16 Jan 2020	25.5	32.0	26.2	21.3	22.0	21.5
17 Jan 2020	24.5	31.0	27.0	22.5	24.0	23.0
18 Jan 2020	24.0	31.0	26.5	22.5	24.5	23.5
19 Jan 2020	18.0	20.2	19.5	19.5	20.5	19.8
20 Jan 2020	17.9	20.0	19.4	19.0	20.1	19.5
Average	**19.8**	**23.2**	**21.1**	**18.6**	**19.8**	**19.2**

Night vs day-night cover

Inner air temperature of night cover (T_2) was always higher than day-night cover (T_3) at 7:00 pm. It was 0.9°C and 0.6°C higher than ambient air temperature and day-night cover, respectively. Day-night cover had 0.3°C higher than ambient air temperature. Similarly, inner air temperaturewas 1.2°C and 0.6°C higher than ambient air temperatureand day-night cover, respectively, at 7:00 am. Day-night cover had 0.6°C higher than ambient air temperature (Table 2).

Table 2. Difference of ambient air and inner temperature of seedbed by polythene cover, Rangpur, Boro 2019-20.

Day	Temperature (°C)					
	7:00 pm			7:00 am		
	Outside polythene cover	Inside polythene cover		Outside polythene cover	Inside polythene cover	
		Night cover	Day-night cover		Night cover	Day-night cover
15 Dec 2019	21.0	23.0	21.3	13.0	14.0	13.0
16 Dec 2019	17.5	18.0	17.5	16.0	17.0	17.0
17 Dec 2019	17.4	18.0	17.0	15.0	17.0	16.0
18 Dec 2019	14.0	14.5	14.3	14.0	15.0	15.0
19 Dec 2019	12.5	13.5	13.4	11.5	12.0	11.7
20 Dec 2019	13.7	14.5	13.9	13.0	13.8	13.5
21 Dec 2019	14.3	14.7	14.5	12.0	13.0	12.7
22 Dec 2019	15.5	15.8	15.6	14.5	15.0	14.8
23 Dec 2019	14.5	15.3	15.0	13.5	14.0	13.8
24 Dec 2019	15.0	15.8	15.4	11.5	13.0	12.7
25 Dec 2019	13.5	15.0	14.8	8.5	9.4	9.0
26 Dec 2019	14.0	16.0	13.0	9.0	11.0	10.5
27 Dec 2019	15.0	15.5	15.2	10.0	12.0	10.4
28 Dec 2019	14.0	14.3	14.1	8.5	10.2	10.0
29 Dec 2019	14.8	15.2	15.0	8.8	10.0	9.0
30 Dec 2019	15.5	16.5	16.0	9.8	10.5	10.0
31 Dec 2019	15.5	17.0	16.0	13.0	14.0	13.5
01 Jan 2020	19.5	20.7	20.2	15.5	17.0	16.0
02 Jan 2020	14.0	15.0	14.6	16.0	16.0	15.5
03 Jan 2020	20.0	22.0	20.0	16.0	20.0	16.3
04 Jan 2020	16.8	17.1	17.0	15.5	16.0	15.8
05 Jan 2020	14.5	14.8	14.6	15.0	15.4	15.0
06 Jan 2020	12.4	13.0	12.6	12.0	13.0	12.8
07 Jan 2020	14.0	15.0	14.5	11.0	12.0	11.5
08 Jan 2020	16.6	17.0	16.8	10.0	11.0	10.6
09 Jan 2020	16.0	17.0	16.5	14.6	15.0	14.8
10 Jan 2020	14.0	15.0	14.5	11.5	12.0	11.8
11 Jan 2020	13.0	14.0	13.6	12.0	12.5	12.3
12 Jan 2020	18.0	20.0	19.0	14.7	15.0	14.8
13 Jan 2020	16.0	16.4	16.2	10.0	11.5	11.5
14 Jan 2020	16.0	17.0	16.5	10.0	11.0	10.8
15 Jan 2020	16.5	17.5	17.0	10.5	16.5	11.0
16 Jan 2020	18.0	19.0	18.5	11.8	12.6	12.0
17 Jan 2020	18.0	19.0	18.5	13.0	13.6	13.4
18 Jan 2020	17.0	18.0	17.5	14.5	15.0	14.8
19 Jan 2020	17.0	18.0	17.7	14.0	15.0	14.7
20 Jan 2020	11.5	12.7	11.1	11.3	12.2	12.0
Average	**15.6**	**16.5**	**15.9**	**12.4**	**13.6**	**13.0**

Seedling strength

Seedling strength was much higher of 1 December seeding than 15 December seeding in both the tested varieties. Normal seedbed (T_4) had higher seedling strength in BRRI dhan88 followed by day-night polythene cover (T_3) at 1 December seeding. It was statistically higher in T_3 followed by T_4 and T_2. Day cover (T_1) had lowest seedling strength in both the varieties. At 15 December seeding, both night cover and day-night cover had higher seedling strength in both the varieties. In the other hand, day cover had also the lowest seedling strength in both the varieties. Seedling strength was lower at 15 December seeding than 1 December seeding might be due to lower Growing degree days (GDD) and prevailing low temperature (below 10°C) for more number of days after seeding up to transplanting (Table 3 and Fig. 1). BRRI dhan89 had higher seedling strength.

Table 3. Effect of polythene cover of seedbed on seedling strength, Boro 2019-20, Rangpur.

Treatment	Seedling strength (mg/cm) D/S: 1 Dec 2019		Seedling strength (mg/cm) D/S: 15 Decr 2019	
	BRRI dhan88	BRRI dhan89	BRRI dhan88	BRRI dhan89
Day cover (**T_1**)	4.40	4.76	1.95	2.01
Night cover (**T_2**)	5.56	6.12	2.51	3.01
Day-night cover (partial opening at both sides) (**T_3**)	5.83	8.56	2.50	4.04
Normal seedbed (control) (**T_4**)	6.14	6.15	0.96	2.88
GDD	749° days		710° days	

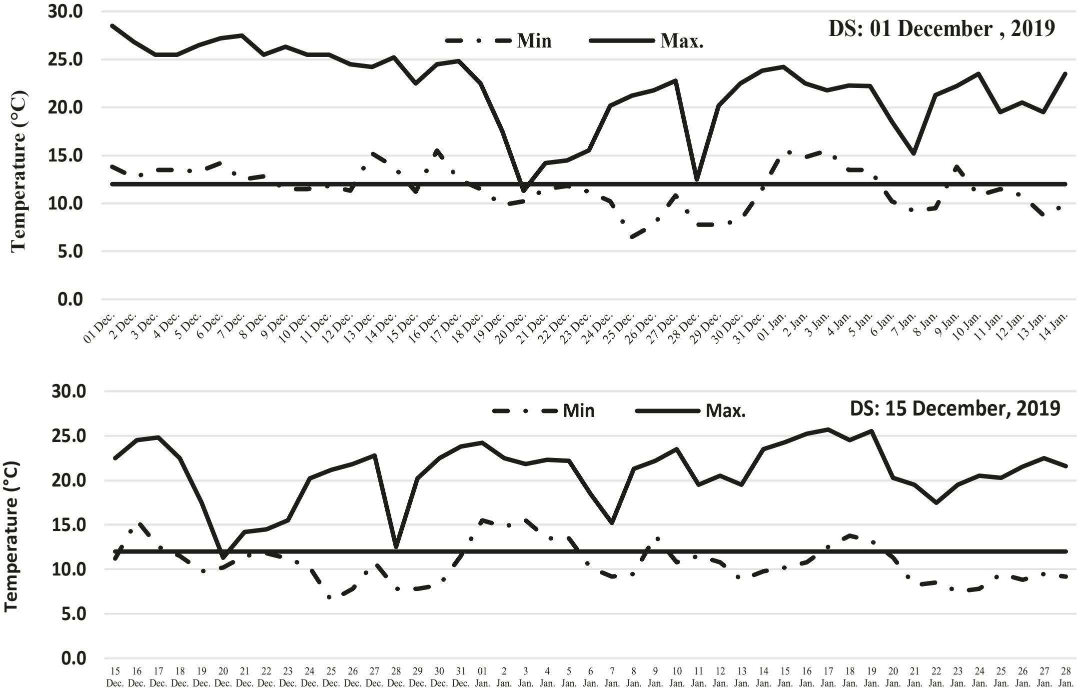

Fig. 1. Minimum and maximum temperature from seeding to seedling uprooting, Boro 2019-20, BRRI RS, Rangpur.

Seedling mortality

Seedling mortality was much higher at 30 January planting than 15 January planting in both the tested varieties. However at 15 January planting,it was higher in control seedbed in both the varieties. Day cover and night cover had similar seedling mortality and day-night (T_3) had the lowest mortality in both the varieties. At 30 January planting, it was also higher in control seedbed in both the varieties followed by day cover and night cover. BRRI dhan89 showed the lowest seedling mortality with day-night cover (T_3). Seedling mortality was higher at 30 January planting than 15 January planting might be due to prevailing low temperature (below 10° C) for more number of days after transplanting (Table 4 and Fig. 2).

Table 4. Effect of polythene cover of seedbed on seedling mortality, Boro 2019-20, Rangpur.

Treatment	Seedling mortality (%) at 25 DAT TP: 15 Jan 2020		Seedling mortality (%) at 25 DAT TP: 30 Jan 2020	
	BRRI dhan88	BRRI dhan89	BRRI dhan88	BRRI dhan89
Day cover **(T_1)**	5.2	3.0	20.2	29.9
Night cover **(T_2)**	5.2	3.0	18.7	20.2
Day-night cover (partial opening at both sides) **(T_3)**	4.5	1.5	17.9	9.0
Normal seedbed (control) **(T_4)**	7.5	7.5	23.9	23.1

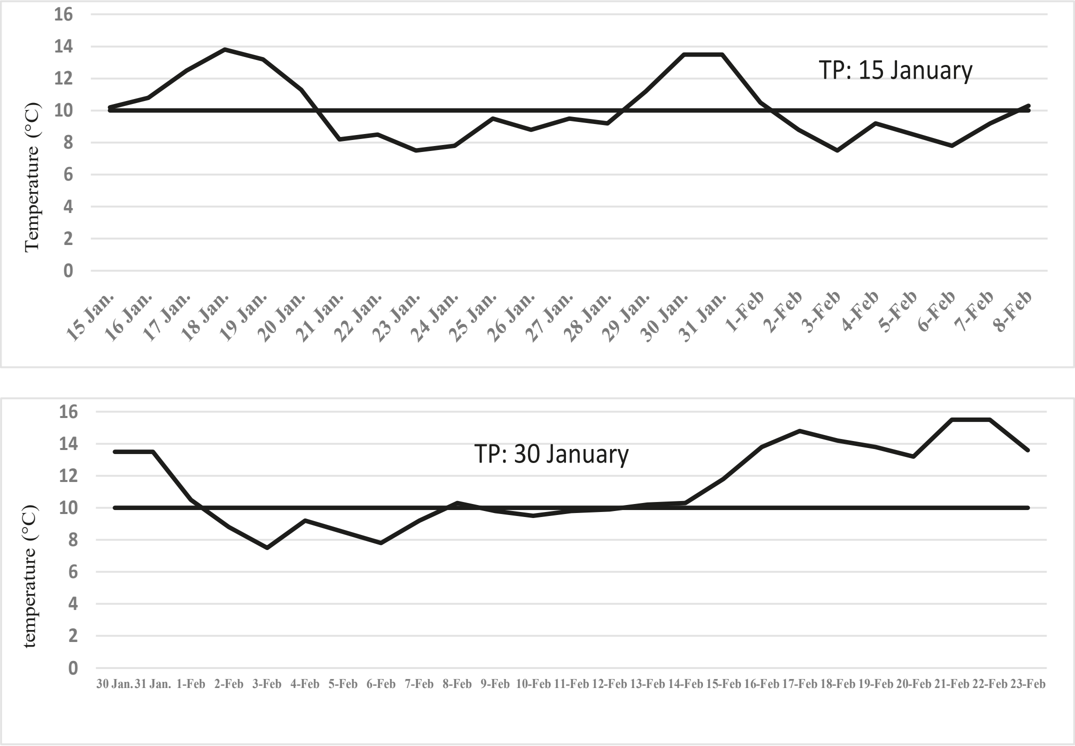

Fig. 2. Minimum temperature from transplanting to 25DAT, Boro 2019-20, BRRI RS, Rangpur.

Seedling height vs plant height

Taller seedling was obtained at 15 January planting but plant height was higher at 30 January planting in both the varieties. In both the plantings, day-night polythene cover treatment (T_3) produced significantly tallest seedling followed by day polythene cover treatment (T_3) in case of both the varieties but it didn't reflect on plant height. Normal seedling (T_4) had the lowest seedling height (Fig. 3). There was no significant difference in plant height at both the plantings in case of both the varieties.

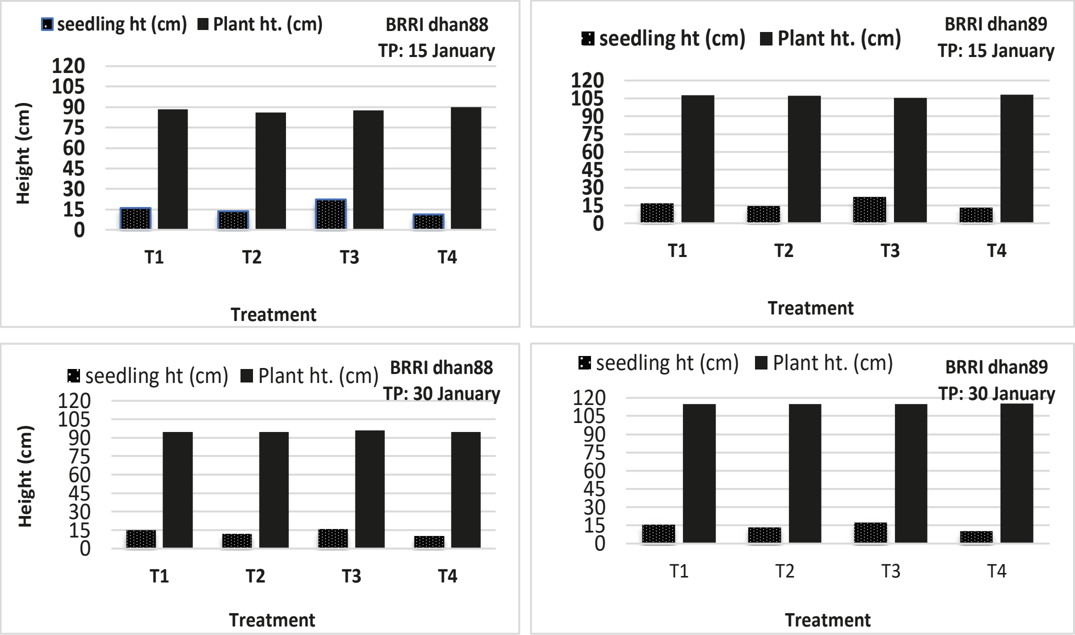

Fig. 3. Effect of polythene cover on seedling height and plant height, Boro 2019-20, BRRI Rangpur.

Tillering pattern

Up to 25 DAT,tiller number was increased very slowly for both the varieties. After 25 DAT, it wasincresed rapidly and reached peak at 65 DATat 15 January planting but it reached peak at 55 DAT of 30 January planting. BRRI dhan88 produced more tiller than BRRI dhan89 at both the plantings. At 15 January planting, BRRI dhan88 produced more tiller with day cover treatment followed by control treatment while day-night cover had the lowest. BRRI dhan89 produced more tiller with night cover treatment followed by no cover treatment but day-night cover had the lowest. At 30 January planting, BRRI dhan88 produced more tiller with day-night and no cover treatment,whereas day cover had the lowest. BRRI dhan89 produced more tiller with day cover treatment followed by night cover treatment,while day-night cover had the lowest (Fig. 4).

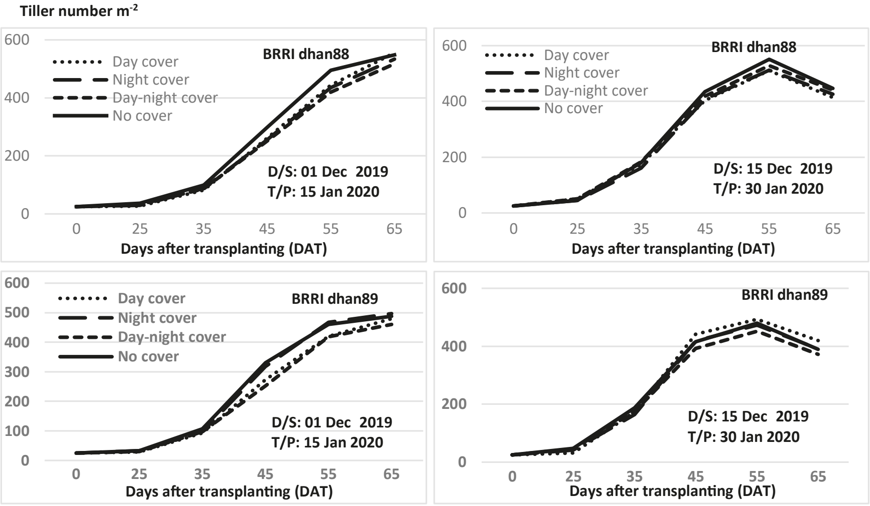

Fig.4. Tillering pattern of BRRI dhan88 and BRRI dhan89 under different polythene cover in seedbed during Boro 2019-20, BRRI RS, Rangpur.

Tiller production rate (no./day)

Tiller production rate was higher at 30 January planting than 15 January up to 45 DAT in both the varieties. At 15 January planting, it increased from 25DAT and sharply increased from 35 – 45 DAT for BRRI dhan88 due to might be increased temperature. It remains steady up to 55 DAT due to reached at maximum tillering stage and then decreased due to tiller mortality. Incase of BRRI dhan89, it wassharply increased from 35 – 45 DAT and then decreased due to tiller mortality. At 30 January planting, tiller production rate gradually increased and reached peak at 45 DAT for both the varieties (Fig. 5). Up to 45 DAT, it was statistically similar in both the varietiers with all treatments.

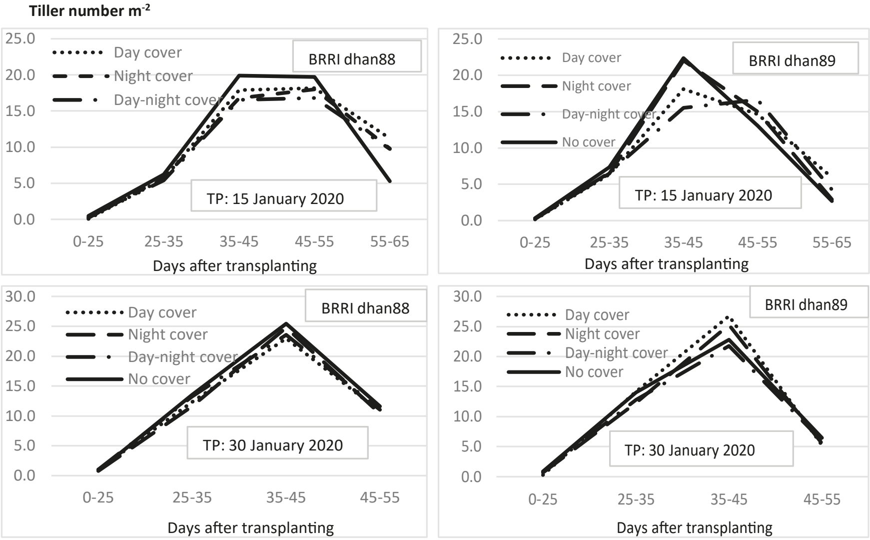

Fig. 5. Tiller production rate of BRRI dhan88 and BRRI dhan89 under different polythene cover in seedbed during Boro 2019-20, BRRI RS, Rangpur.

Productive tiller (%)

Polythene covering treatments of rice seedling didn't vary significantly in both the plantings but BRRI dhan88 had higher productive tiller rate at 30 January planting(Table 5). Although, tiller number was higher in T_1, T_2 and T_4 than T_3 but productive tiller rate was satistically similar in both the varieties, plantings due to might be production of early tiller caused by higher seedling strength.

Table 5. Effect of polythene cover of on productive tiller, Boro 2019-20, Rangpur.

Treatment	Productive tiller (%) TP: 15 Jan 2020		Productive tiller (%) TP: 30 Jan 2020	
	BRRI dhan88	BRRI dhan89	BRRI dhan88	BRRI dhan89
Day cover **(T_1)**	53.5	55.5	69.3	59.1
Night cover **(T_2)**	53.6	55.9	67.8	58.4
Day-night cover (partial opening at both sides) **(T_3)**	53.5	55.4	66.8	55.5
Normal seedbed (control) **(T_4)**	54.2	56.6	63.9	55.0
$Lsd_{0.05}$	ns	ns	ns	ns

Panicle number vs grain yield

Both the varieties produced higher number of panicles at 30 January planting and BRRI dhan88 produced more number of panicle with all the treatments (Table 6). There was no significant difference in grain yield at both the planting in both the varieties. BRRI dhan89 produced higher grain yield due to more number of grains per panicle and it was higher at 30 January planting (Table 6 and 7). Day-night polythene cover treatment (T_3) reduces growth duration by 2-3 days over other treatments might be due to higher seedling strength caused by no contact with fog and cold wave.

Table 6. Effect of polythene cover treatment in seedbed on yield and ancillary characters at 15 January planting, Boro 2019-20, BRRI RS, Rangpur .

Treatment	Panicle m^{-2}	TGD (days)	Yield (t ha^{-1})	Panicle m^{-2}	TGD (days)	Yield (t ha^{-1})
		BRRI dhan88			BRRI dhan89	
Day cover **(T_1)**	297	154	6.67	266	166	7.35
Night cover **(T_2)**	286	153	6.62	278	166	7.52
Day-night cover (partial opening at both sides) **(T_3)**	277	152	6.50	255	163	7.63
Normal seedbed (control) **(T_4)**	297	153	6.78	276	166	7.68
$Lsd_{0.05}$	18.7	ns	ns	16.79	ns	ns

Table 7. Effect of polythene cover treatment in seedbed on yield and ancillary characters at 30 January planting, Boro 2019-20, BRRI RS, Rangpur.

Treat	Panicle m^{-2}	TGD (days)	Yield (t ha^{-1})	Panicle m^{-2}	TGD (days)	Yield (t ha^{-1})
	BRRI dhan88			BRRI dhan89		
Day cover **(T_1)**	354	151	6.99	307	164	7.99
Night cover **(T_2)**	347	150	7.18	299	164	8.08
Day-night cover (partial opening at both sides) **(T_3)**	353	149	7.42	293	161	8.29
Normal seedbed (control) **(T_4)**	352	150	7.24	303	163	7.96
$Lsd_{0.05}$	ns	ns	ns	ns	ns	ns

CONCLUSION

Day-night polythene cover treatment (T_3) produces less tiller than day or night polythene cover and no polythene cover treatment but productive tiller (%) was satistically similar among the treatments for both the varieties and planting dates. Moreover, day-night polythene cover treatment (T_3) reduces growth duration by 2-3 days than the other treatments. Although, there was no significant difference in grain yield among the treatments in both the planting dates but day-night polythene cover (partial opening at both sides) produces taller and stronger seedling that helps farmers to easy uprooting and transplanting in time. Also, treatment T_3 is farmers friendly over T_1 and T_2 because it is hassle free, a few labour consuming (cost effective) and risk free that has the potentiality to produce higher seedling strength.

REFERENCES

Ali, M G, REL Naylor and S Matthews. 2006. Distinguishing the effects of genotype and seed physiological age on low temperature tolerance of rice (*Oryza sativa* L.), *Experimental Agriculture*, 42 (3): 337-349

Alvarado, R and S Hernaiz. 2007. Antecedentes General essobreel arroz en Chile, in Arroz Manejo Tecnológico, R. Alvarado, Ed., of Boletín INIA, Instituto de Investigaciones Agropecuarias INIA, Centro Regional de Investigación Quilamapu, Chillán, Chile, 162:179

Baruah,A R, N Ishigo-Oka and M Adachi. 2009. Cold tolerance at the early growth stage in wild and cultivated rice. Euphytica, 165(3): 459-470

Farrell, T C, K M Fox, R L Williams, S Fukai and L G Lewin. 2006. Minimising cold damage during reproductive development among temperate rice genotypes. II. Genotypic variation and flowering traits related to cold tolerance screening, *Australian Journal of Agricultural*, 57(1): 89-100

Fujino, K, H Sekiguchi and T Sato. 2004. Mapping of quantitative trait loci controlling low-temperature germinability in rice (Oryza sativa L.), *Theoretical and Applied Genetics*, 108 (5): 794-799

Nishiyama, I. 1995. Type of damage due to cool weather and the relevant research. *In*: Science of the rice plant. Vol. 2. *Food and Agriculture Policy research Centre*. Tokyo, Japan, pp 769-798

Shimono, H, T Hasegawa and K Iwama. 2002. Response of growth and grain yield in paddy rice to cool water at different growth stages, *Field Crops Research*, 73 (2/3): 67-79

Tajma, K. 1995. Factors and mechanism of chilling injury. *In*: Science of the rice plant. Vol. 2. *Food and Agriculture Policy research Centre*. Tokyo, Japan, pp 798-799

Bangladesh Rice J. 26 (1): 69-78, 2022, doi.org/10.3329/brj.v26i2.66593

Variability and Genetic Gain Prediction for Maintainer Line Improvement of Hybrid Rice in Bangladesh

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ABSTRACT

Assessment of genetic variability in the nursery of the breeding population is essential for crop improvement successfully. Thirteen maintainer lines of hybrid rice were evaluated to estimate the level of their genetic diversity and heritability of grain yield influencing parameters in the transplanted Aman 2020 season. The studied traits were days to 50% flowering, total effective tillers plant^{-1}, plant tallness (cm), length of flag leaf (cm), breadth of flag leaf (cm), panicle size (cm), filled grains panicle^{-1}, unfilled grains panicle^{-1}, growth duration (days) and grain yield (tha^{-1}). Coefficient of variation (genotypic and phenotypic) was noticed high for most traits that revealed high variability among the studied genotypes. Broad-sense heritability (h^2_{bs}) was high in all traits except flag leaf breadth. Analysis of the cluster and its mean comparison showed that cluster 2 (i.e. BRRI 11B, BRRI 99B, IR 79125B and IR 79156B) represented the best agronomic traits and yield potentials. Therefore, selection of genotypes with valuable attributes from cluster 2 will be considered for maintainer line improvement programmes. The use and estimation of predicted genetic gain will provide a visionary insight of the future genotypes produced after the crossing of the genotypes under study.

Key words: Maintainer line, genetic advance, heritability, predicted genetic gain, hybrid rice

INTRODUCTION

Ever growing and dense population allied with yield ceiling of the staple food rice has become a curse for Bangladesh. Here rice takes up 75% (BBS, 2017) to 78% (Kabir *et al.*, 2020) of the total cropped area. Commercial exploitation of heterosis in rice was the weapon to feed the people of China and 55% rice area was used to produce 66% of overall rice production (Virmani *et al.*, 1998). Promising and potential hybrids out yielded modern and best rice varieties (conventional varieties) by 15-20% and 20-30%, respectively (Yuan, 1998). Polygenic trait like grain yield is impacted by environments where the genotypes grow and is estimated by the nature and degree of genetic variation (Selvaraj *et al.*, 2011). Variability among the genotypes expressed as genetic divergence used for gene pool broadening and needs trustworthy heritability estimates to design breeding strategy with high efficiency (Akinwale *et al.*, 2011). Broad sense-heritability provides knowledge about the overall variability accounted for by genotypic effect (Allard, 1960). Maintainer lines (B lines) are the key genotypes that are used to supply yield boosting genes to the female parent (A line) during new female line development. Maintainer lines (B lines) are mainly developed using B×B crossing method (Virmani *et al.*, 1998). Hybrid rice breeders have to select new and better B×B

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combinations to develop new elite maintainers for CMS lines multiplication and as well hybrid rice production. Heritability, genetic distance, genotypic coefficient of variation (GCV), phenotypic coefficient of variation (PCV); environmental coefficient of variation (ECV), coefficient of variation (CV), genetic advance (GA), clustering of genotypes help the breeder to effectively select cross combinations e.g. B×B cross combination selection in case of maintainer line improvement. Previous studies suggested that superior rice genotypes should measure and achieve genetic gain with sensory perception and grain quality attributes (Anacleto *et al.*, 2015). Empirical evaluation plus genetic prediction will need to be complemented by the future plant breeders (Cooper *et al.*, 2014). So the present investigation was conducted to estimate genetic variability, heritability, genetic advance and predicted genetic gain of the studied maintainer lines.

MATERIALS AND METHODS

During T. Aman, 2020 (July - December, 2020) season; 13 maintainer lines (enlisted in Table 2) of hybrid rice were assessed in three replications using RCB design at the research field (West Byed) of Bangladesh Rice Research Institute (BRRI). The experimental plots were monocrop area where rice is grown throughout the year. Chemical fertilizers @ 194-82-52-60 kg/ha for urea-MoP-TSP-gypsum, respectively were used in the field to ensure nutrient supply to plants. Complete urea was administered in three splits i.e. 10 days after transplantation (DAT), 30 DAT and 45 DAT. At the time of final land preparation, complete TSP, MoP, and gypsum were applied. Thirty-days-old seedlings were transplanted with a spacing of (25 cm × 15 cm). Data from each replication were obtained from randomly selected 10 plants. Data collection on 50% flowering (days), total effective tillers hill^{-1}, plant height (cm), length of flag leaf (cm), breadth of flag leaf (cm), filled grains panicle^{-1}, length of panicle (cm), unfilled grains panicle^{-1}, growth duration (days) and grain yield (tha^{-1}).

STATISTICAL ANALYSIS

Variance analysis was conducted with the collected data from the RCB design of this experiment using the STAR Version: 2.0.1 (Statistical Tool for Agricultural Research) software for genetic divergence and cluster analysis. Mean sum of squares were utilized to assess genetic parameters e.g. genotypic variance (σ_g^2), phenotypic variance (σ_p^2) environmental variance (σ_e^2), Genotypic coefficient of variation (GCV), genetic advance (GA), coefficient of variation (CV), phenotypic coefficient of variation (PCV) and environmental coefficient of variation (ECV) to display variability among maintainer line genotypes. Multivariate cluster analysis using the method of Agglomerative Cluster Analysis (Ward's method) was done. Ten yield and yield-contributing traits were used for genetic divergence and cluster analysis.

Total variation of each character was divided into non-genetic and genetic parts and GCV, PCV, ECV, %CV were assessed in line with Burton (1952) and Sharma (1988):

$$\sigma_{\mathrm{g}}^2 = \frac{\mathrm{MS}_{\mathrm{g}} + \mathrm{MS}_{\mathrm{e}}}{\mathrm{r}}, \ \sigma_{\mathrm{e}}^2 = \mathrm{MS}_{\mathrm{e}}, \ \sigma_{\mathrm{p}}^2 = \sigma_{\mathrm{g}}^2 + \sigma_{\mathrm{e}}^2$$

where σ_p^2 = phenotypic variance, σ_g^2 = genotypic variance, and σ_e^2 =environmental variance and MS_g = mean squares of genotypes, MS_e= mean squares of error, and r = number of blocks .

$$\%\mathrm{CV} = \frac{\sqrt{MS_g}}{\bar{X}} \times 100, \ \mathrm{PCV} = \frac{\sqrt{\sigma_{\mathrm{p}}^2}}{\bar{X}} \times 100, \ \mathrm{GCV} = \frac{\sqrt{\sigma_{\mathrm{g}}^2}}{\bar{X}} \times 100, \ \mathrm{ECV} = \frac{\sqrt{\sigma_{\mathrm{e}}^2}}{\bar{X}} \times 100$$

Where, $\bar{X}$=grand mean for each measured traits . h_{bs}^2 = Broad sense heritability which is expressed as the proportion of the genotypic variance (σ^2g) to phenotypic variance (σ^2p) ratio and was assessed as stated by Burton (1952). Genetic advance (GA) was projected by the technique defined by Fehr (1987) as $GA = K \times \sqrt{\sigma_p^2} \times h_{bs}^2$, where at 5% pressure of selection the value of constant K is 2.06, ($\sqrt{\sigma_p^2}$) = phenotypic standard deviation and h_{bs}^2 = broad sense heritability, GA = genetic advance was also assessed as proportion of the average. We have used **RStudio Version 1.1.463** to calculate the predicted genetic gain/year. The expected or predicted genetic gain/year was estimated as:

$$\Delta G = \frac{i\, r\, \sigma_A}{t}$$

Here, ΔG = predicted genetic gain/year, i = selection intensity (mean deviance of carefully chosen entries in units of σ_A (phenotypic standard deviation), r = accuracy of selection, σ_A = standard deviation of breeding values (Falconer and Mackay, 1996) or genetic standard deviation ($\sqrt{\text{additive genetic variance}}$), and t = time or duration per breeding cycle (Yunbi *et al.* 2017). In genomic selection (GS) analysis, r = the correlation between TBVs (true breeding values) and GEBVs (genomic-estimated breeding values), while in case of phenotypic selection, r = h_{ns}^2 and thus $\Delta G = \frac{i h \sigma_A}{t}$ (Bassi *et al.*, 2016; Heffner *et al.*, 2010; Meuwissen, 2003). In our experiment, we used the expected accuracy, r = $\sqrt{1 - \frac{PEV}{V_g}}$ (where, V_g = Genetic variance, and PEV = unexplained part of V_g by the predictions) that is supported by Pszczola *et al.* 2012, Hayes *et al.* 2009, VanRaden 2008. In this article, genotypes were presumed to be unrelated.

RESULT AND DISCUSSION

Genetic variability

Diverse breeding materials with high genetic variability are a prerequisite to guide a breeding program towards success. Understanding the variability and magnitude in maintainer lines (B Line) is crucial as it delivers the foundation of parent selection for B x B improvement in hybrid rice breeding. Table 3 presents the genetic parameters and mean squares of 13 maintainer lines of hybrid rice are presented in Table 3.

Table 1. Quantitative traits related mean square with genetic parameters of 13 maintainer lines in T. Aman, 2020.

Traits	MS_g	σ^2_e	σ^2_g	σ^2_p	Mean	GCV	PCV	ECV	%CV	H^2_{bs} (%)	GA
50%F	94.92***	1.09	31.28	32.37	77.82	7.19	7.31	1.40	12.52	96.63	11.33
GD	61.37***	1.09	20.09	21.18	102.51	4.37	4.49	1.06	7.64	94.87	8.99
Etill	5.09***	0.56	1.51	2.07	7.87	15.61	18.27	7.11	28.65	72.94	2.16
Yield	0.72***	0.11	0.20	0.32	3.45	13.02	16.29	3.31	24.59	63.86	0.74
FGP	3041.3***	151.39	963.30	1114.69	127.77	24.29	26.13	118.49	43.16	86.42	59.44
UFGP	2721.63***	36.89	894.91	931.80	58.28	51.33	52.38	63.30	89.51	96.04	60.39
PL	9.29***	0.10	3.06	3.16	21.93	7.98	8.11	0.47	13.89	96.73	3.54
PH	223.35***	2.18	73.72	75.90	87.81	9.78	9.92	2.48	17.02	97.13	17.43
FLL	39.62***	1.60	12.67	14.27	34.96	10.18	10.81	4.58	18.00	88.78	6.91
FLB	0.04*	0.02	0.01	0.02	1.58	5.74	9.87	1.02	12.78	33.84	0.11

Legends: 50%F=days to 50% flowering, Etill=total effective tillers hill^{-1}), PH=plant height (cm), FLL=flag leaf length (cm), FLB=flag leaf breadth (cm), PL=panicle length (cm), FGP=filled grains panicle^{-1}), UFGP=unfilled grains panicle^{-1}), GD=growth duration (days), Yield=grain yield (tha^{-1}), MS_g = mean squares of genotypes, σ_p^2 = phenotypic variance, σ_g^2 =genotypic variance, σ_e^2 =environmental variance genotypic coefficient of variation (GCV), phenotypic coefficient of variation (PCV), environmental coefficient of variation (ECV), coefficient of variation (%CV), h_{bs}^2 =broad sense heritability, GA= genetic advance, * and ***= significant at the 5% and the 0.1% level.

ANOVA exhibited significant ($p < 0.001$) differences for all the studied characters in the maintainer lines except flag leaf breadth that was significant ($p < 0.05$) marginally (Table 1). The significant variations detected among the maintainer lines for all the traits influenced the presence of intrinsic genetic variability among the studied maintainers. Akter *et al.* (2019) described the presence of genetic difference among hybrid rice genotypes. Breeding programs related to the betterment of yield requires genetic variation in the selected mating populations to effectively select and achieve yield upgrading (Ndukauba *et al.*, 2015 ; Idahosa *et al.*, 2010). Percentage of CV relates the relative quantity of variability in the traits of crop plant (Sharma, 1988). The highest percentage of CV obtained by the unfilled grains panicle^{-1} followed by filled grains panicle^{-1}, effective tiller hill^{-1} and yield (tha^{-1}) (Table 1). These results indicated that the unfilled grains panicle^{-1} followed by filled grains panicle^{-1}, effective tiller hill-1 and yield (tha^{-1}), respectively, had greater quantities of accessible genetic variability amongst the studied maintainer lines. It also implied the bigger prospect of yield improvement in choosing these traits compared to rest of the traits (Ndukauba *et al.*, 2015; Eid, 2009). On the contrary, the lowest coefficient of variation was noted for growth duration, days required to flower 50% and flag leaf breadth exhibited low utilizable genetic variability that has less potential of satisfactory advancement in selecting these traits contrasted with other traits. The phenotypic variance (σ_p^2) of the studied traits was separated into genotypic variance (heritable) and environmental variance (non-heritable) constituents (Table 1). Genotypic variances were greater than their related environmental variances in all the traits, except flag leaf breadth which was negligible (Table 1). This specified that the total variation was contributed mainly by the component of genotypic variation in the considered traits. The maximum PCV was found for the unfilled grains panicle^{-1} followed by filled grains panicle^{-1}, effective tiller hill^{-1}, and yield while the smallest PCV was recorded for growth duration, panicle length, flag leaf breadth and plant height. High PCV specifies the presence of a bigger scope of choice for the characters of interest, which was determined by the quantity of variability exist (Naik *et al.*, 2020; Khan *et al.*, 2009). Thus, ample potential for selecting the filled grains panicle^{-1}, effective tiller hill^{-1}, yield and flag leaf length among the tested maintainer lines is predicted. In contrast, there was a minor scope of choice for growth duration, panicle length, flag leaf breadth, plant height as a consequence of low variability. Diverse quantitative traits exhibited genetic variability in plants and estimated by GCV. Unfilled grains panicle^{-1}, filled grains panicle^{-1}, effective tiller hill^{-1}, yield (tha^{-1}) and flag leaf length showed the highest amount of GCV, respectively. Growth duration, flag leaf breadth, 50% flowering and panicle length contrariwise, showed the least amount of GCV (Table 1). The existence of utilizable genetic variability for different traits is indicated by high GCV, which can simplify selection effectively (Naik *et al.*, 2020; Yadav *et al.*, 2009). The range obtained for environmental coefficient of variation

(ECV) was 0.47 (panicle length) to 118.49 (filled grain panicle^{-1}). Though estimates for phenotypic coefficient of variation (PCV) were higher than those estimates for genotypic coefficient of variation (GCV), they were close; inferring that trait expression is governed mainly by lines compared to environment and phenotypic value based selection is therefore feasible. Whereas, a large inequality between GCV and PCV estimations for flag leaf breadth specified a greater amount of environmental regulation for these traits. Variation governed by polygene can be phenotypic, genotypic or environmental and the relative estimates of GCV, PCV and ECV for a trait provides knowledge about the degree of variability (Ndukauba *et al.*, 2015; Nausherwan *et al.*, 2008).

HERITABILITY ESTIMATES

Heritability estimates offer a vision into the degree of genetic regulation to express individual characteristics and phenotypical reliability of breeding value prediction (Ndukauba *et al.*, 2015). High heritability estimate of a trait indicates low environmental effect in the detected variation (Eid, 2009). h^2_{bs} only shows whether there is adequate genetic variation in any population, which infers about the population response to selection pressure (Gatti *et al.*, 2005; Milatovic *et al.*, 2010; Ullah *et al.*, 2012). Heritability of the studied traits ranged from 33.84% (flag leaf b) to 97.13% (plant height). Heritability of all the traits except plant height was above 60% (Table 1). GCV, PCV, ECV and heritability results of this experiment explained the existence of considerable extent of genetic variation in these traits to permit parent assortment for the development of better maintainer line. These traits should be under special consideration when choosing parents of maintainer line improvement programme. To achieve more effective character selection, heritability supplemented with genetic advance is more suitable than heritability on its own (Ullah *et al.*, 2012). For most of the traits, high h^2_{bs} was reported in the current study, but were associated with low genetic advance except unfilled grains panicle^{-1} (Genetic advance= 60.39) and filled grains panicle^{-1} (Genetic advance= 59.44) (Table 1). High heritability connected with high genetic advance for a certain trait resulted due to the actions of additive gene and offers an effective situation for selection (Rashid *et al.*, 2017; Gyawali *et al.*, 2018; Ndukauba *et al.*, 2015; Tazeen *et al.*, 2009).

Genetic divergence of the maintainer lines

Narrow distance indicates the most similar genotype pairs and long distance shows diverse genotype pairs. The longest Euclidean distance was 7.0 (between IR79125B and BRRI50B) and the shortest distance was 1.8 (between BRRI97B and BRRI35B) (Table 2). The genotype pairs that exhibited long distance will be used for new elite parental line development (Table 2).

Table 2. Euclidean distances of maintainer line genotypes under study.

	BRRI 10B	BRRI 11B	BRRI 35B	BRRI 48B	BRRI 50B	BRRI 7B	BRRI 97B	BRRI 99B	IR 105687B	IR 105688B	IR 58025B	IR 79125B	IR 79156B
BRRI 10B	0.0	4.1	2.3	2.8	4.2	3.3	2.5	3.7	3.9	3.7	4.3	4.5	5.2
BRRI 11B	4.1	0.0	3.5	4.4	5.0	4.3	3.2	2.9	6.0	5.0	5.1	4.8	3.8
BRRI 35B	2.3	3.5	0.0	2.6	3.8	2.3	1.8	3.0	4.2	3.6	4.9	5.4	5.3
BRRI 48B	2.8	4.4	2.6	0.0	4.3	2.5	2.3	4.2	4.6	4.1	5.0	6.0	6.2
BRRI 50B	4.2	5.0	3.8	4.3	0.0	4.6	4.4	5.8	5.0	4.6	4.9	7.1	6.7
BRRI 7B	3.3	4.3	2.3	2.5	4.6	0.0	2.7	4.2	4.7	4.1	5.1	5.7	6.3
BRRI 97B	2.5	3.2	**1.8**	2.3	4.4	2.7	0.0	2.8	4.1	3.4	4.6	5.2	5.1
BRRI 99B	3.7	2.9	3.0	4.2	5.8	4.2	2.8	0.0	5.6	4.5	5.9	4.7	4.6
IR 105687B	3.9	6.0	4.2	4.6	5.0	4.7	4.1	5.6	0.0	2.3	3.2	5.3	5.6
IR 105688B	3.7	5.0	3.6	4.1	4.6	4.1	3.4	4.5	2.3	0.0	3.8	5.1	5.5
IR 58025B	4.3	5.1	4.9	5.0	4.9	5.1	4.6	5.9	3.2	3.8	0.0	4.1	4.3
IR 79125B	4.5	4.8	5.4	6.0	**7.1**	5.7	5.2	4.7	5.3	5.1	4.1	0.0	3.4
IR 79156B	5.2	3.8	5.3	6.2	6.7	6.3	5.1	4.6	5.6	5.5	4.3	3.4	0.0

Three clusters were formed at distance coefficient 7 having 6, 4 and 3 entries in cluster 1, cluster 2, and cluster 3, respectively (Fig 1). Cluster 1 showed the moderate value for yield, filled grain panicle-1, panicle length, flag leaf breadth; and the lowest value for 50% flowering date, growth duration, effective tiller hill^{-1} and unfilled grains panicle-1 (Table 3). Cluster 2 occupied the highest value for 50% flowering date, growth duration, yield, filled grains panicl^{e-1}, flag leaf length, plant height; moderate value for effective tiller hill^{-1}, unfilled grains panicle^{-1}; and the lowest value for flag leaf breadth. Cluster 3 contained the highest value for effective tiller hill^{-1}, unfilled grains panicle^{-1} and flag leaf breadth; moderate value for 50% flowering date, growth duration, flag leaf length and the lowest value for yield, panicle length, plant height and filled grains panicle^{-1}. Similar method of Euclidean distance based clustering was applied to select parent for hybridization programme of rice crop by breeders (Adhikary *et al*., 2018, Nitesh *et al*., 2014).

Table 3. Cluster mean of ten traits utilized in the grouping of 13 maintainer lines in T. Aman, 2020.

Traits	Cluster 1	Cluster 2	Cluster 3
50%F	73.80	81.60	80.90
GD	100.23	106.00	102.47
Etill	7.05	7.85	9.57
Yield	3.50	3.70	3.00
FGP	130.85	137.75	108.33
UFGP	33.85	57.53	96.80
PL	21.03	24.02	20.83
PH	84.45	98.05	81.00
FLL	32.53	39.30	34.03
FLB	1.62	1.50	1.63

Legends: 50%F=days to 50% flowering, Etill=total effective tillers hill-1, PH=plant height (cm), FLL=flag leaf length (cm), FLB=flag leaf breadth (cm), PL=panicle length (cm), FGP=filled grains panicle^{-1}, UFGP = unfilled grains panicle^{-1}, GD = growth duration (days), Yield=grain yield (tha^{-1})

The cluster 1 contained six lines viz. BRRI 10B, BRRI 35B, BRRI 48B, BRRI 50B, BRRI 7B and BRRI 97B. Four maintainers BRRI 11B, BRRI 99B, IR 79125B and IR 79156B formed cluster 2. The smallest cluster contained only three maintainers i.e. IR 105687B, IR 105688B and IR 58025B. Maintainers in the same cluster had more similarity than the maintainers in different cluster.

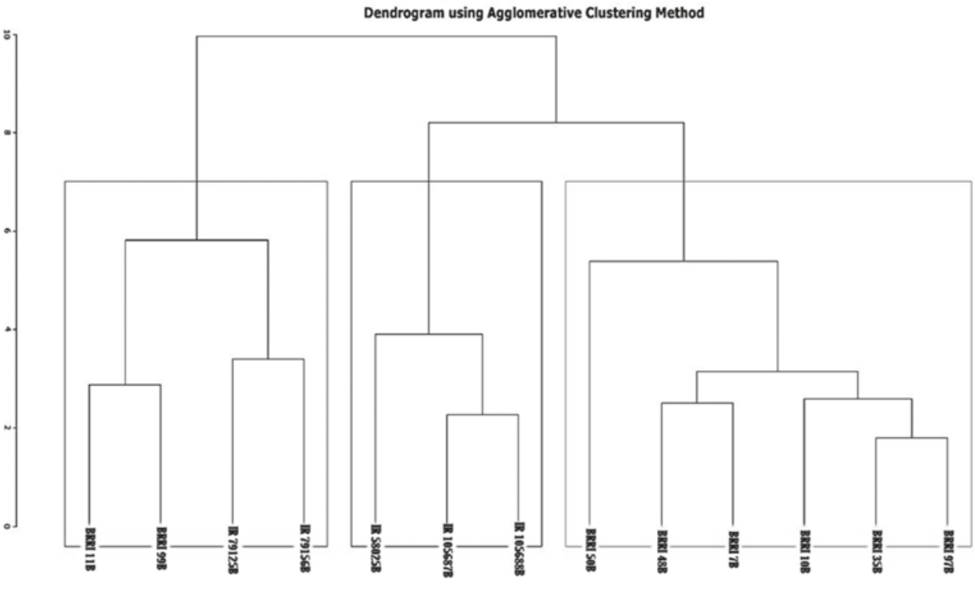

Fig. 1. Dendrogram showing clusters of 13 maintainer lines of hybrid rice genotypes obtained using a set of 10 characters.

Predicted genetic gain:

At present hybrid rice division, BRRI needs six years to complete a breeding cycle for B×B improvement. About 0.084 tha^{-1} $year^{-1}$ genetic gains can be achieved from the genotypes used in this experiment at 30% selection intensity and six year breeding cycle length. At the same time, 10% selection intensity and 6 year breeding cycle length will produce 0.117 tha-1year-1genetic gain (Table 4). The population size will be needed to increase 10-fold to double genetic gain and rising selection intensity from 0.1% to 0.01% only raises the projected gains by 20% (approximately) (Yunbi *et al.* 2017). We're concentrating on shortening the breeding cycle using field rapid generation advance. If the length of breeding cycle is reduced compared to the present; it will escalate the genetic gain for the studied genotypes also and table 4 presents the conditions.

Table 4. Predicted genetic gain ($tha^{-1}year^{-1}$) in different situations for the studied maintainers.

Breeding cycle length (Year)	Selection intensity	Predicted genetic gain ($tha^{-1}year^{-1}$)	Breeding cycle length (Year)	Selection intensity	Predicted genetic gain ($tha^{-1}year^{-1}$)
3	5%	0.275	3	10%	0.234
4	5%	0.206	4	10%	0.176
5	5%	0.165	5	10%	0.141
6	5%	0.138	6	10%	0.117

CONCLUSION

Four promising maintainer lines (viz BRRI 11B, BRRI 99B, IR 79125B and IR 79156B) were selected for the transplanted Aman rice (Wet season). The best maintainer lines will be further used in cyclic breeding to develop new elite maintainer lines. Genotype clustering into several clusters (3) advocates moderate genetic variation among genotypes to warrant improvement through breeding for transplanted Aman season.

REFERENCES

Akinwale, A G, G Gregorio, F Nwilene, B O Akinyele, S A Ogunbayo and A C Odiyio. 2011. Heritability and correlation coefficient analysis for yield and its components in rice (*Oryza sativa* L.). *African Journal of Plant Science*, 5: 207-212.

Akter, A, M J Hasan, M A Latif, M U Kulsum, P L Biswas, M H Rahman, R R Majumder, L F Lipi, M R Quddus, F Akter and A Ara. 2019. Genetic Variability, heritability, correlation and path coefficient studies for yield and yield components of some promising rice hybrids. *Bangladesh Rice Journal*, 23 (2): 27-34, DOI: doi.org/10.3329/brj.v23i2.48245

Allard, R W. 1960. Principles of plant breeding. 1st edn. John Wiley and Sons Inc, New York.

Anacleto, R, R P Cuevas, R Jimenez, C Llorente, E Nissila, R Henry and N Sreenivasulu. 2015. Prospects of breeding high-quality rice using postgenomic tools. Theoretical and Applied Genetics, 128: 1449–1466.

Bassi F M, A R Bentley, G Charmet, R Ortiz and J Crossa. 2016. Breeding schemes for the implementation of genomic selection in wheat (Triticum spp.). Plant Science, 242: 23–36.

BBS (Bangladesh Bureau of Statistics). 2017. Yearbook of Agricultural Statistics-2016. Bangladesh Bureau of Statistics (BBS), Statistics and Informatics Division, Ministry of Planning, Government of the People's Republic of Bangladesh, Dhaka, Bangladesh.

Burton, G W. 1952. Qualitative inheritance in grasses. Vol. 1. p. 277-283. Proceedings of the 6th International Grassland Congress, Pennsylvania State College. 17-23 August. Pennsylvania State College, Pennsylvania, USA.

Cooper, M, C D Messina, D Podlich, L R Totir, A Baumgarten, N J Hausmann, D Wright and G Graham. 2014. Predicting the future of plant breeding: complementing empirical evaluation with genetic prediction. *Crop and Pasture Science*, 65: 311–336.

Eid, M H. 2009. Estimation of heritability and genetic advance of yield traits in wheat (Triticum aestivum L.) under drought conditions. *International Journal of Genetics and Molecular Biology*, 1(7): 115-120.

Falconer, D S and T F C Mackay. 1996. Introduction to Quantitative Genetics, 4th edn. London: Longman Group Ltd.

Fehr, W I. 1987. Principles of Cultivar Development. Vol. 1. 548 p. Macmillan, New York, USA.

Gatti, I, F L Anido, C Vanina, P Asprelli, and E Country. 2005. Heritability and expected selection response for yield traits in blanched asparagus. *Genetics and Molecular Research*, 4(1): 67-73.

Gyawali, S, A Poudel and S Poudel. 2018. Genetic variability and association analysis in different rice genotypes in mid hill of western Nepal. *Acta Scientific Agriculture*. 2(9): 69-76.

Hayes, B J, P J Bowman, A C Chamberlain, K Verbyla and M E Goddard. 2009. Accuracy of genomic breeding values in multi-breed dairy cattle populations. Genetics Selection Evolution. 41: 51.

Heffner, E L, A J Lorenz, J L Jannink and M E Sorrells. 2010. Plant breeding with genomic selection: gain per unit time and cost. *Crop Science*, 50: 1681–1690.

Idahosa, D O, J E Alika, and A U Omoregie. 2010. Genetic variability, heritability and expressed genetic advance as indices for yield and yield components selection in cowpea (Vigna unguiculata (L.) Walp.). *Academia Arena*, 2 (5): 22-26.

Khan, A S M M R, M Y Kabir, and M M Alam. 2009. Variability, correlation, path analysis of yield and yield components of pointed gourd. *Journal Agricultural Rural Development*, 7 (1-2): 93-98.

Meuwissen, T H E. 2003. Genomic selection: the future of marker assisted selection and animal breeding. In: FAO Workshop on Marker-Assisted Selection: A Fast Track to Increase Genetic Gain in Plant and Animal Breeding? Session II: MAS in Animals. Turin, Italy, 17-18 October, 54–59. https://scholar.google.com/scholar?cluster=6325434300706126096&hl=en&as

Milatovic, D, D Nikolic and D Durovic. 2010. Variability, heritability and correlation of some factors affecting productivity in peach. *Horticultural Science (Prague)*, 27 (3): 79-87.

Naik, M V K, M Arumugam Pillai, and S Saravanan. 2020. Genetic variability studies in F1 rice (Oryza sativa L.) hybrids for yield and quality traits. *The Journal of Phytopharmacology*. 9(6): 453-458.

Nausherwan, N N, M S Ghulam, M Khali, S Qamar and S Akhtar. 2008. Genetic variability, correlation, path analysis studies in garden pea (Pisum sativum L.). *Journal of Agricultural Research*, 36 (1): 333-340.

Ndukauba, J, G E Nwofia, P I Okocha, and E E Ene-Obong. 2015. Variability in Egusi-Melon Genotypes (Citrulluslanatus [Thumb] Matsum and Nakai) in derived Savannah environment in South-Eastern Nigeria. *International Journal of Plant Research*, 5 (1): 19-26. DOI:10.5923/ j.plant. 20150501.04.

Nitesh, K, P K Singh, A Vaishampayan, Rajesh Saini, Mukh Ram, Aparajita Singh and N K Singh. 2014. Genetic Divergence Analysis in Rice under Irrigated Conditions. *Indian Journal of Plant Genetic Resources*. 27(3): 246-250.

Pszczola, M, T Strabel, H Mulder and M Calus. 2012. Reliability of direct genomic values for animals with different relationships within and to the reference population. *Journal of Dairy Science*, 95: 389–400.

Rashid, M M, M Nuruzzaman, L Hassan and S N Begum. 2017. Genetic variability analysis for various yield attributing traits in rice genotypes. *Journal of Bangladesh Agricultural University*. 15(1): 15-19.

Kabir M S, M U Salam, A K Islam, M A Sarkar, M A Mamun, M C Rahman, B Nessa, M J Kabir, HB Shozib, M B Hossain, A Chowdhury. 2020. Doubling rice productivity in Bangladesh: A way to achieving SDG 2 and moving forward. *Bangladesh Rice Journal*. 24 (2): 1-47.

Selvaraj, I C, P Nagarajan, K Thiyagarajan, M Bharathi and R Rabindran. 2011. Genetic parameters of variability, correlation and path coefficient studies for grain yield and other yield attributes among rice blast disease resistant genotypes of rice (Oryza sativa L.). *African Journal of Biotechnology*, 10: 3322-3334.

Sharma, J R. 1988. Statistical and biometrical techniques in plant breeding. 432 p. New Age International Limited Publishers, New Delhi, India.

Tazeen, M, K Nadia, and N N Farzana. 2009. Heritability, phenotypic correlation and path coefficient studies for some agronomic characters in synthetic elite lines of wheat. *Journal of Food, Agriculture and Environment*, 7 (3-4): 278-282.

Ullah, M Z, M J Hassan, A Z M K A Chowdhury, A I Saki, and A H M A Rahman. 2012. Genetic variability and correlation in exotic cucumber (Cucumis sativus L.) varieties. *Bangladesh Journal of Plant Breeding and Genetics*, 25 (1): 17-23.

VanRaden, P. 2008. Efficient methods to compute genomic predictions. *Journal of Dairy Science*. 91: 4414–4423.

Virmani, S S, P J Jachuck, S D Chatterjee and M I Ahmad. 1998. Opportunities and challenges of developing hybrid rice technology for rainfed lowland and Boro ecosystem. *In*: Rainfed Rice foor Sustainable Food Security (S. K. Mohanty, ed). Cuttack, India. 533-62 pp.

Yadav, Y C, B B Sanjay Kumar, and S K Dixit. 2009. Genetic variability, heritability and genetic advance for some traits in Cucumber. *Indian Journal of Agricultural Research*, 8: 51-57.

Yuan, L P. 1998. Hybrid rice breeding in China. *In*: Advances in Hybrid Rice Technology (Virmani SS, Siddiq EA, Muralidharan K, editors). Proc. 3rd Intl. Symp. on Hybrid Rice. held on 14-16 November 1996, Hyderabad, India. 27-33 pp.

Yunbi, Xu, L Ping, C Zou, L Yanli, ChuanxiaoXie, X Zhang, M Boddupalli, Prasanna, S Michael and Olsen. 2017. Enhancing genetic gain in the era of molecular breeding. *Journal of Experimental Botany*, 68 (11): 2641–2666. DOI:10.1093/jxb/erx135

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