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Effect of Water Deficit Stress on Yield Performances in Wet Seeded Rice

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ABSTRACT

Worldwide fresh water scarcity and labour unavailability in agriculture are driving researchers and farmers to find management strategies that will increase water productivity and reduce labour requirement. Wet seeding instead of transplanting rice greatly reduces the labour requirement for crop establishment, while use of alternate wetting and drying (AWD) instead of continuous flooding reduces irrigation input. However, the safe threshold for irrigating wet seeded rice (WSR) at different crop stages has not been investigated. Therefore, experiment was conducted to determine the effects of different degrees of water stress during different crop growth stages on yield performance of WSR. This was done in greenhouse experiment in the 2011 wet season 2011 at the International Rice Research Institute, Los Baños, Philippines. In the experiment, water stresses were applied by withholding irrigation until soil water tension increased to 10, 20 or 40 kPa (kilo pascal) at 10 cm below the soil surface. Soil water tension was measured using 30 cm long guage tensiometer installed with the center of the ceramic cup. The stresses were applied during three crop stages: 3-leaf (3L) to panicle initiation (PI), PI to flowering (FL), and FL to physiological maturity (PM). The experiment also included a continuously flooded (CF) treatment. The number of drying events ranged from 8-12 during 3L-PI, 6-10 during PI-FL and 6-10 during FL-PM. There was a consistent trend for a decline in the number of irrigations and irrigation input with increasing irrigation threshold, and thresholds of 20 and 40 kPa resulted in significantly lower input than with CF. There were consistent trends for lower grain yield as the level of water deficit stress increased, and imposition of stresses of 20 and 40 kPa at any or all three stages significantly reduced grain yield compared with CF. There was a trend for the reduction in grain yield to be greater when the stresses were imposed at all three stages compared with a single stage, but the differences were not significant. There was a consistent trend for irrigation water productivity (WPi) to decrease as the irrigation threshold increased, with significantly lower values for a 40 kPa threshold at any stage, in comparison with CF. This was because the decline in water input to the pots was less than the decline in yield as the threshold increased. The results suggest that the optimum threshold for irrigation of WSR is 10 kPa during the vegetative and grain filling stages, and that the soil should be kept at close to saturation during PI-FL.

Key words: Water productivity, soil water tension, wet season, physiological maturity

INTRODUCTION

In most of Asia, irrigated rice is manually transplanted into puddled soil, and the fields are continuously flooded for most of the season until shortly before harvest. However, peak period labour scarcity has greatly increased the cost of transplanting and often results in delayed crop establishment. Therefore, there is a growing interest in direct seeded rice because of its low labour requirement. There are three basic forms of direct seeding – water seeding, dry seeding and wet seeding (Kumar and Ladha, 2011). Wet seeding involves puddling of the soil prior to sowing pre-germinated seed by manual broadcasting or line seeding (using a manually pulled drum seeder, Rashid *et al.*, 2009) onto the saturated soil surface. Like irrigated, puddled transplanted rice (PTR), wet seeded rice (WSR) requires large amounts of water for puddling and to keep the field continuously flooded after establishment. Furthermore, WSR may require more irrigation water than PTR as it takes for longer time in the main field (Cabangon *et al.*, 2002). Assuming the increasing scarcity of water for agriculture

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together with the need to increase production to feed growing populations, there has been a lot of effort to identify methods for reducing irrigation water input to rice while maintaining yield (Guerra *et al.*, 1998; Bouman *et al.*, 2005; Humphreys *et al.*, 2010). However, lowland rice varieties are highly sensitive to soil drying, with yields generally declining once the soil starts to dry below saturation. In their review, Bouman and Tuong (2001) found yield reductions of 0-12% when the soil was kept at saturation instead of flooded, and of 10-40% when soil tension in the root zone was allowed to increase to 10 to 30 kPa. However, safe alternate wetting and drying (AWD) water management has proven to be a successful and practical means of reducing irrigation input to PTR by 10-40% while maintaining yield (Bouman and Tuong, 2001). With safe AWD, the field is flooded to a shallow depth (~5 cm) for about two weeks after transplanting, after which irrigation is managed to ensure that the soil water tension at 15 cm depth does not exceed about 10 kPa. The same threshold is applied throughout the duration of the crop, except during the heading/flowering period when keeping the soil saturated/flooded is recommended (Bouman *et al.,* 2005). While a threshold of 10 kPa is safe in a range of soil types, there are situations where higher thresholds of 20 to 60 kPa throughout the season did not cause a loss of yield (eg Kukal *et al.*, 2005; Bueno *et al.*, 2010; Sudhir-Yadav *et al.*, 2011). The safe threshold depends on many factors including genotype, soil type, frequency of drying events, evaporative demand, and growth stage.

The effect of water deficit stress at different crop stages has been the subject of many drought studies, primarily targeted at rainfed, lowland rice. These studies typically involved imposing high levels of water deficit stress by withholding irrigation for many days until the crop was stressed to the degree of complete leaf rolling (soil tension in excess of 500 kPa, Wopereis *et al.*, 1996), or until there was significant leaf death. This was then followed by well-watered conditions to maturity. These studies showed that periods of drought delayed phenological development, but much more so (by up to about three weeks) when the stress was applied during the vegetative phase (Woopereis *et al.*, 1996; Castillo *et al.*, 2006; Davatgar *et al.*, 2009). In these studies, applying stress during the reproductive stage (panicle initiation, booting, flowering) was more detrimental to grain yield than applying stress during the early vegetative stage, while applying the stress during mid tillering was also detrimental. However, while it is generally agreed that the period during anthesis is especially sensitive to drought stress, the review of Bouman and Tuong (2001) found no systematic trend for greater yield reduction when drought was imposed in certain development stages compared with other developmental stages. The results of De Datta *et al,* (1973) (in Bouman *et al.*, 2001) showed that different cultivars had different responses to drought stress timing and intensity. With soil drying to 50 kPa, some cultivars showed more yield reduction with drought imposed during the vegetative stage than in the reproductive stage, some showed the reverse and some showed higher yields with drought in the vegetative stage than in the well-watered control.

In irrigated rice culture, the usual goal of water saving irrigation management is to avoid stressing the crop to the degree that yield is reduced, while minimizing irrigation input. The degree to which the soil can be allowed to dry during different crop phases without affecting yield, and whether there is a cumulative effect of low levels of water deficit stress across all crop stages are not well understood. Furthermore, in contrast with the situation for transplanted rice, reports on the response of WSR to soil drying below saturation are lacking. Therefore, the objectives of the study were: to understand the effect of threshold level on number and amount of irrigation and effect of water stress on yield and yield components.

MATERIALS AND METHODS

The research was carried out in the greenhouse at the International Rice Research Institute

(IRRI), Los Baños, Philippines (14°11′N, 121°15′E), from July to November 2011. Rice was grown on a silty clay loam soil (28% sand, 34% silt, 39% clay) in 20 cm diameter, 25 cm high polyvinyl chloride pots. Bulk soil was obtained from the IRRI experimental station, homogenized well with water and added to the pots to a depth of 20 cm. The soil was re-puddled using an electrical stirrer before wet seeding. The soil was slightly acidic, with medium levels of organic C, available P, exchangeable Ca and K, and total N (Table 1).

The experiment was designed to evaluate the effects of different levels of water stress at different growth stages. There were three levels of water stress (10, 20 and 40 kPa soil water tension) applied at one of three growth stages: 3 leaf (3L) to panicle initiation (PI), PI to flowering (FL), and FL to physiological maturity (PM) (Table 2). In addition there was a control treatment, which was continuously flooded (CF), and three treatments, which had stresses of 10, 20 or 40 kPa during all the three stages. Thus, there were 13 water management treatments in a randomized complete block design.

Implementation of CF involved topping the pots up daily to a pond water depth of 2 to 3 cm. For the stress treatments, irrigation was applied whenever soil water tension increased to the threshold value (10, 20 or 40 kPa), with water added in two doses, topping up to a depth of 2 cm each time, to ensure that the soil was fully saturated to depth. Soil tension was measured using 30-cm long tensiometers installed in 4 replicate pots of each treatment. The tensiometers were installed 4 cm to the side of the centre of the plant rows, and the middle of the ceramic cup was 10 cm below the soil surface. All pots of a given treatment were irrigated when the average of the four monitored pots reached the threshold value. All pots were kept continuously flooded at all stages other than during the stage when the 10, 20 or 40 kPa stresses were being applied.

Table 2. Water management treatments.

Treatment	Irrigation threshold during each crop stage[a] (kPa)		
	3L to PI	PI to FL	FL to PM
CF-CF-CF	CF	CF	CF
10-CF-CF	10	CF	CF
20-CF-CF	20	CF	CF
40-CF-CF	40	CF	CF
CF-10-CF	CF	10	CF
CF-20-CF	CF	20	CF
CF-40-CF	CF	40	CF
CF-CF-10	CF	CF	10
CF-CF-20	CF	CF	20
CF-CF-40	CF	CF	40
10-10-10	10	10	10
20-20-20	20	20	20
40-40-40	40	40	40

[a]3L=3 leaf stage, PI=panicle initiation, FL=flowering, PM=physiological maturity, CF=continuously flooded.

After the final puddling, the soil was allowed to settle for 1 d. A solution of 27 ml of fertilizer containing muriate of potash, diammonium phosphate and zinc sulphate was poured onto the soil surface 24 h before seeding, giving a basal fertilizer application rate of 40 kg K_2O ha^{-1}, 41 kg P_2O_5 ha^{-1}, 30 kg N ha^{-1} and 5 kg Zn ha^{-1}. Urea was top-dressed at maximum tillering, PI and heading at 50, 50 and 30 kg N ha^{-1}, respectively, and muriate of potash (40 kg ha^{-1}) was also applied at PI. The top dressing was done immediately prior to irrigation.

The rice variety NSIC Rc222 was pre-germinated by soaking the seeds for 24 h, draining, then incubating for 24 h prior to sowing. NSICRc222 was selected for its high yield potential under non-continuously flooded

Table 1. Chemical properties of the soil used in the greenhouse experiment.

pH (1:5) in H_2O)	Organic C (g. 100g^{-1})	Olsen P (mg.kg^{-1})	Exch. Ca (meq.100g^{-1})	Exch. K (meq.100g^{-1})	Total N (g. 100g^{-1})
6.0	1.56	39.3	15.3	0.90	0.17

conditions. The seeds were placed on the saturated soil surface on 20 July 2011. Twelve pre-germinated seeds were sown in a single 20 cm long row along the diameter of each pot. The plants were thinned to eight plants per pot after establishment. The pots were kept weed free by hand weeding as needed.

Insects were well-controlled by applying insecticides as needed against whorl maggot, leaf and plant hoppers, stem borer, and rice bugs.

Air temperature, relative humidity and solar radiation were measured hourly using an automatic weather station in the greenhouse near the middle of the experiment. The same parameters were also measured outdoors at the IRRI lowland farm weather station, located among rice fields about 1 km from the greenhouse.

All data were analyzed by analysis of variance (ANOVA) using GenStat V.14.1. The interaction between water stress treatment and growth stage was also analyzed using a factorial design with three levels of water stress (10, 20 and 40 kPa) and three stages of stress application (3L - PI, PI - FL, FL - PM). The comparison of treatment means was made by the least significant difference (LSD) at the 5% level of probability (p=0.05).

RESULTS AND DISCUSSIONS

Weather

Temperature in the greenhouse generally ranged from 30-35°C, with daily minimum temperature only a couple of degrees lower than maximum temperature (Fig. 1a). Maximum air temperature inside the greenhouse was usually slightly higher than the outside air temperature. However, minimum temperature inside the greenhouse was higher than the outside temperature by several degrees. Relative humidity (%) in the greenhouse was usually about 10% lower than that of the outside (Fig. 1b). Solar radiation in the greenhouse was usually much lower inside the greenhouse than that of the outside, by about 50%, except on rainy, overcast days (Fig. 1c). There was strong linear relationship between solar radiation inside and outside the greenhouse (Fig. 1d).

Effect of water stress treatment on the frequency and amount of irrigation

The soil in the water stress treatments dried rapidly after each irrigation (Figs 2a-c, 3a-c). For example, during 3L-PI, a period of about 42 d, the 10 kPa treatment received 11 irrigations, an average of 1 irrigation every 3.8 d. The frequency of irrigation was only slightly less in the 20 and 40 kPa treatments. The frequency of irrigation within stress treatments was higher during FL-PM than the other two periods, with the 10 kPa receiving an average of one irrigation every 3.3 d.

Fig. 2. Soil water tension in the 10, 20 and 40 kPa water stress treatments from three leaf to panicle initiation (a), panicle initiation to flowering and (b) from flowering to physiological maturity in the greenhouse (c) at IRRI, Los Banos, Philippines during July to November 2011. Data are means of four replicates.

Fig. 3. Soil water tension in the 10-10-10 (a), 20-20-20 and (b) 40-40-40 (c) kPa water stress treatments from three leaf to physiological maturity in the greenhouse at IRRI, Los Banos, Philippines during July to November 2011. Data are means of four replicates.

This is consistent with the findings of the field experiments of Sudhir-Yadav *et al.* (2011) with both puddled transplanted and dry seeded rice on a clay loam soil with a deep water table. They found a large reduction in the number of irrigations in going from CF to an irrigation threshold of 20 kPa, but only small differences in the number/frequency of irrigations as the threshold increased from 20 to 40 to 70 kPa. This was partly due to rains, but even in dry spells there were only small delays in drying from 20 to 40 to 70 kPa.

During crop establishment, the total number of irrigations and the volume of water added were the same for all the treatments. Imposing water stress at all stages greatly reduced the number of irrigations in comparison with the CF treatment, but the effect on the volume applied

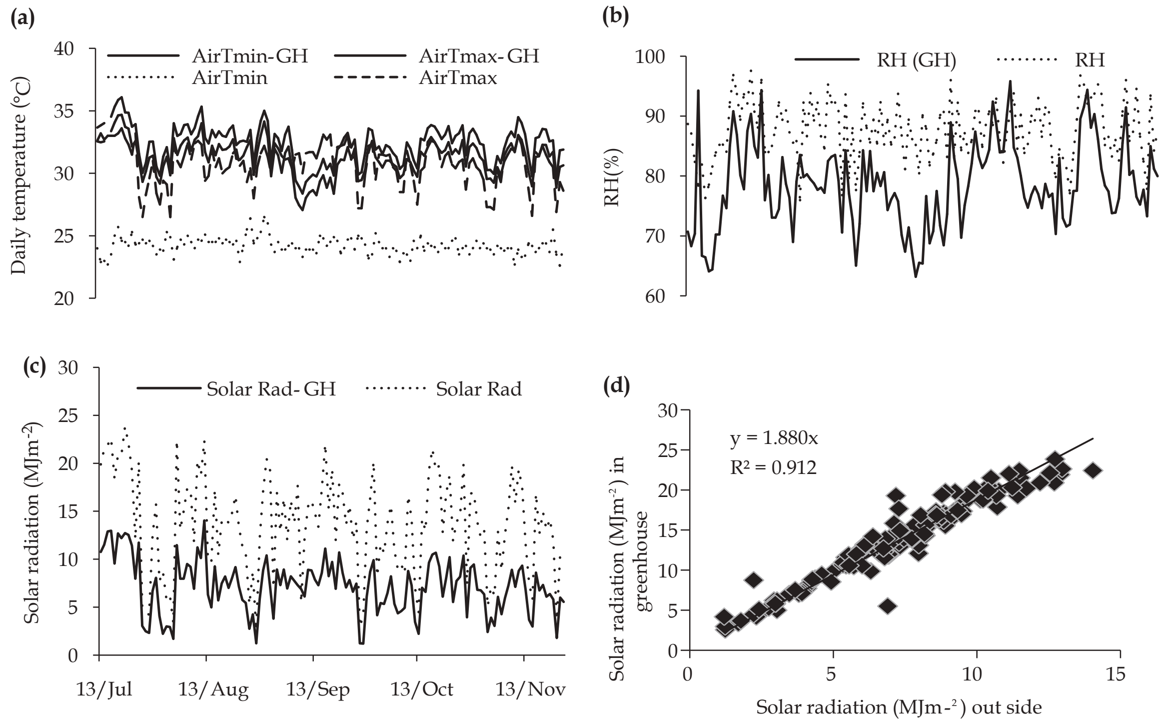

Fig. 1. Daily maximum and minimum temperature (a), daily relative humidity (b), daily solar radiation (c), and the relationship between solar radiation inside and outside the greenhouse (GH) (d) at IRRI, Los Baños, Philippines in 2011.

was negligible (Table 3). During 3L-PI there were small differences between some treatments in the volume applied, but with no meaningful trends. During PI-FL, the treatments with thresholds of 20 and 40 kPa at 3L-PI and/or PI-FL received significantly less water than the CF treatment. During FL-PM, all stress treatments received less water than the CF treatment, regardless of when the stress was applied, except 10-CF-CF. The net result was that total water input to the treatments with 20 or 40 kPa irrigation thresholds at some or all stages was significantly lower (by about 20%) than input to the CF treatment. There was a consistent trend for higher total water application with a 10 kPa threshold than with higher thresholds, with significantly lower water input in 40-CF-CF compared with 10-CF-CF (by 14%) in CF-CF-40 compared with CF-CF-10 (by about 15%), and in 20-20-20 and 40-40-40 compared with 10-10-10 (by 15-20%). The effect of the irrigation treatments on irrigation amount was relatively small in comparison with the findings of many field experiments (eg Belder *et al.*, 2004; Sudhir-Yadav *et al.*, 2011). However, the main causes of the irrigation reduction in field experiments were usually reduced seepage and percolation losses (Bouman *et al.,* 2005), whereas, there was no drainage or seepage from the pots. In non-draining pots, irrigation is only needed to replace water lost by evapotranspiration. In treatments with higher irrigation thresholds, the soil dries to a greater degree before irrigation, and therefore a larger amount of water is needed to return the soil to saturation. In non-draining pots differences in irrigation amount thus reflect differences in evaporation and transpiration. The lack of a significant reduction in irrigation input with a threshold of 10 kPa compared with CF is in contrast with the results of field experiments, which show significant irrigation reduction when changing from CF to safe AWD (threshold of ~10 kPa), even for part of the season (Bouman and Tuong, 2001).

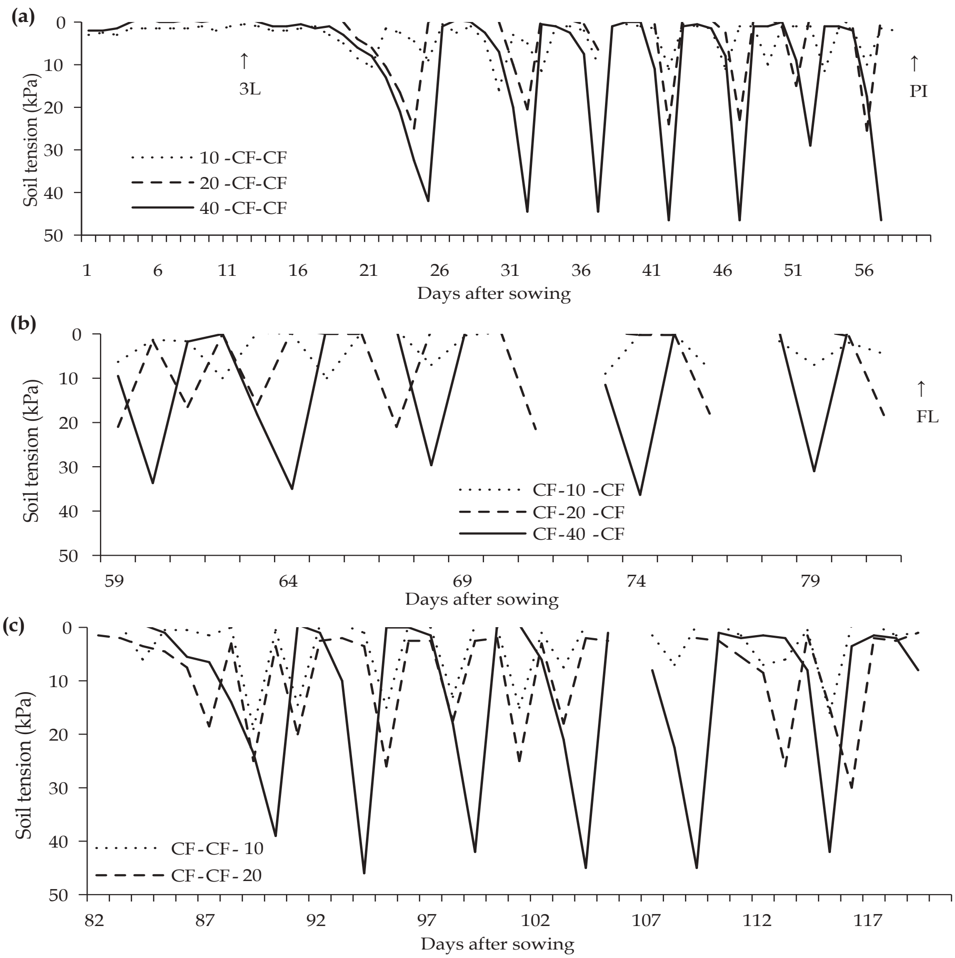

Fig. 2. Soil water tension in the 10, 20 and 40 kPa water stress treatments from three leaf to panicle initiation (a), panicle initiation to flowering and (b) from flowering to physiological maturity in the greenhouse (c) at IRRI, Los Banos, Philippines during July to November 2011. Data are means of four replicates.

Effect of water stress treatment on grain yield and yield components

There was a consistent trend for declining grain yield with increasing water stress during all stages, and stresses of 20 and 40 kPa applied during any or all of the three stages significantly reduced yield (Table 4). There was a consistent trend for a greater reduction in yield when the stresses of 20 and 40 kPa were applied at all three stages than during a single stage, but the differences were never significant. A stress of 10 kPa did not have a significant effect on yield, except when applied during PI-FL only. The lack of yield reduction with a threshold of 10 kPa is consistent with the findings of Sharma (1997) in drums in a glasshouse, and of De Datta *et al.* (1973b) who found only small effects of drying to 10 kPa tension on grain yield, even when the accumulated number of days without water was as many as 38. However, Sharma (1989) found a small (8%) but significant yield reduction with a threshold of 10 kPa in the field in one of two years. In the present greenhouse experiment, the 10-10-10 treatment received 41 irrigations, meaning that the pots were not ponded for about 41 days (or significant parts thereof). In their review, Bouman and Tuong (2001) reported that yields were reduced by

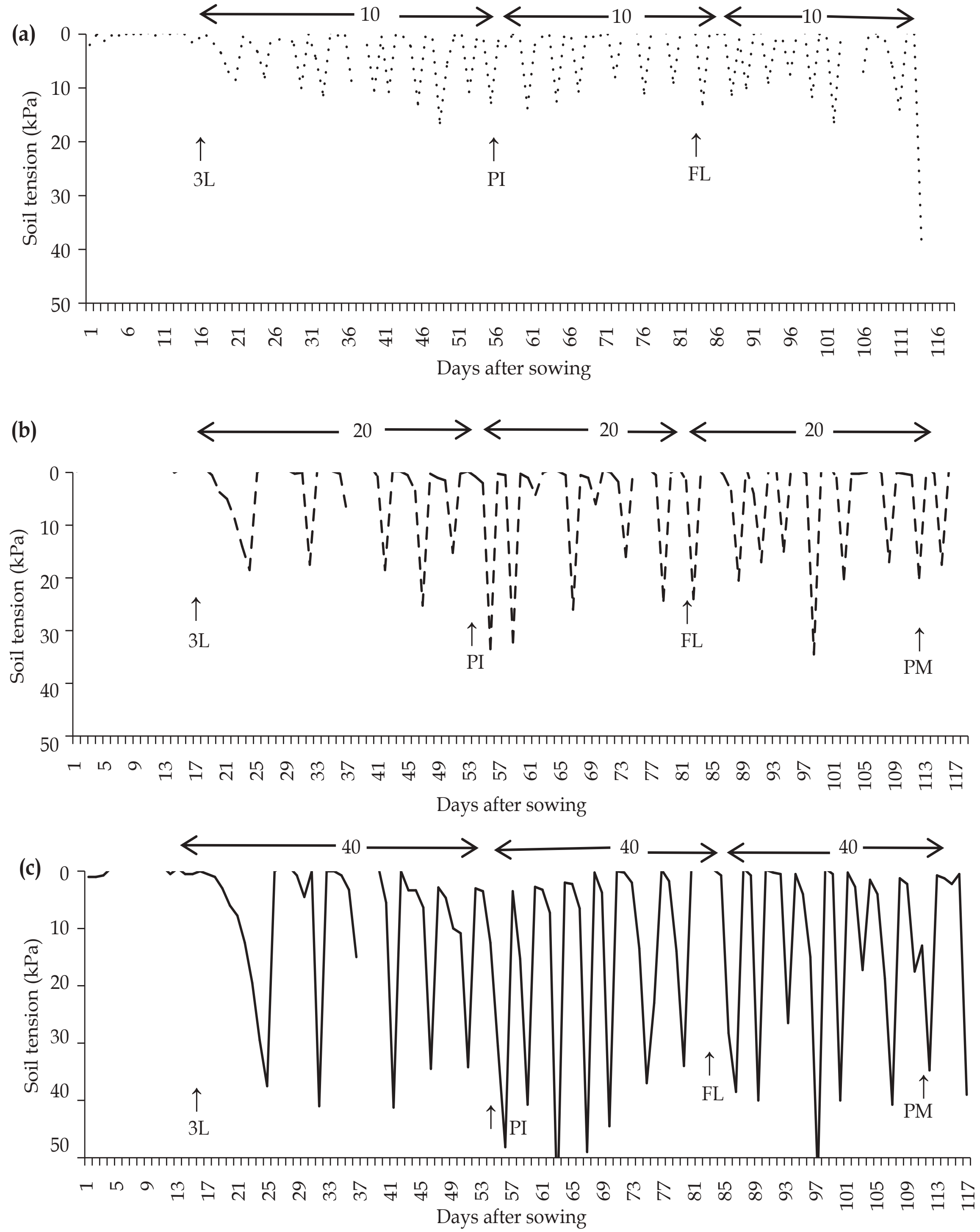

Fig. 3. Soil water tension in the 10-10-10 (a), 20-20-20 (b) and 40-40-40 (c) kPa water stress treatments from three leaf to physiological maturity in the greenhouse at IRRI, Los Banos, Philippines during July to November 2011. Data are means of four replicates.

Table 3. Effect of irrigation threshold on the number of irrigations (No.) and volume applied during each crop stage and the whole growing season.

Treatment	Crop establishment		3L-PI		PI-FL		FL-PM		Whole season	
	No.	Volume (lit.)	No.	Volume (lit.)	No.	Volume (lit.)	No.	Volume (lit.)	No.	Volume (lit.)
CF-CF-CF	12	1.2	29.3	9.1	21.5	10.8	33.0	17.0	95.8	38.0
10-CF-CF	12	1.2	11	9.7	18.5	9.3	31.3	15.8	72.8	36.1
20-CF-CF	12	1.2	10	11.9	18.0	7.3	28.8	12.6	68.8	32.9
40-CF-CF	12	1.2	8	9.0	18.0	8.1	28.3	12.6	66.3	30.9
CF-10-CF	12	1.2	32.8	9.9	10.0	10.6	31.8	11.7	87.5	33.5
CF-20-CF	12	1.2	30.0	8.8	8.25	8.1	30.5	12.7	80.8	30.8
CF-40-CF	12	1.2	30.3	9.1	6.0	7.3	29.5	12.9	77.8	30.6
CF-CF-10	12	1.2	28.5	9.5	21.8	11.0	10	13.2	72.3	34.9
CF-CF-20	12	1.2	29.3	9.0	22.0	9.4	10	12.2	73.3	31.8
CF-CF-40	12	1.2	29.0	8.4	18.5	8.5	6	9.2	65.5	27.2
10-10-10	12	1.2	12	10.9	7	9.2	10	13.7	41.0	34.9
20-20-20	12	1.2	9	8.7	7	8.5	8	10.1	36.0	28.6
40-40-40	12	1.2	9	9.2	6	8.3	8	11.2	35.0	29.8
$LSD_{0.05}$			2.32	1.63	1.9	2.1	2.55	2.09	4.8	3.9

3L=3 leaf stage, PI=panicle initiation, FL=flowering, PM=physiological maturity, CF=continuously flooded.

Table 4. Grain yield, yield components, straw yield and harvest index (HI) as affected by water stress imposed at different growth stages compared with continuous flooding.

Treatment [a]	Pan pot^{-1} (no.)	Floret pan^{-1} (no.)	Floret fertility (%)	1000 grain wt (g)	Grain yield (dry) (g pot^{-1})	Straw yield (g pot^{-1})	HI
CF-CF-CF	24	94	86.6	21.5	41.2	50.6	0.44
10-CF-CF	18	105	90.3	21.3	34.2	48.6	0.41
20-CF-CF	19	79	86.9	21.2	27.8	43.9	0.38
40-CF-CF	19	63	86.9	21.4	21.7	44.9	0.33
CF-10-CF	20	82	85.0	21.5	30.3	45.8	0.39
CF-20-CF	22	79	85.0	20.9	27.7	41.8	0.39
CF-40-CF	20	80	77.0	21.1	25.1	42.5	0.37
CF-CF-10	22	97	88.5	21.7	41.7	53.2	0.43
CF-CF-20	20	89	85.7	21.7	33.3	44.4	0.42
CF-CF-40	20	73	83.3	20.1	24.7	41.9	0.36
10-10-10	19	103	84.7	21.8	35.3	51.1	0.40
20-20-20	16	91	85.0	21.4	26.4	40.1	0.39
40-40-40	14	84	86.2	21.2	21.4	37.4	0.36
$LSD_{0.05}$	5	ns	ns	ns	8.0	ns	0.06

[a]Data were analyzed by RCBD.

10-40% when soil water potentials in the root zone were allowed to reach 10 to 30 kPa. A threshold of 40 kPa during a single stage (3L-PI, FL-PM) and at all three stages significantly reduced yield by 39-48% in comparison with the flooded control, and yields with 40 kPa were also lower than yields of all treatments with 10 kPa thresholds at one or all stages, but similar to yields with 20 kPa thresholds. Castillo *et al.* (2006) found similar yield reductions with severe stress applied during part of the period PI-FL or part of FL-PM. In a field experiment, De Datta *et al.* (1973b) found 30 and 40% yield reductions with a 15 kPa threshold for two varieties (IR20 and MI-48), and no further reduction with a threshold of 40 kPa.

There was a consistent trend for lower panicle density in all stressed treatments than in CF, with significantly lower values when stresses were applied during the 3L-PI stage or during all the three stages. When the stresses were applied at all stages, panicle density declined as the threshold increased, with significantly lower values in 40-40-40 than 10-10-10. Panicle density is determined by both tiller production during the vegetative stage and by tiller mortality after maximum tillering. Sudhir-Yadav *et al.* (2011) found greater tiller mortality as irrigation threshold increased from daily to 20, 40 and 70 kPa following a non-stressed period up to maximum tillering. There was a consistent trend for fewer florets per panicle with 20 and 40 kPa stresses than with CF during any or all the three stages, but with no significant differences. Floret fertility and 1000 grain weight were not affected by irrigation treatment and there were no consistent trends. Floret fertility is determined during the period of pollen cell formation (a couple of weeks prior to FL) and during anthesis. There were no significant differences in floret fertility, which had a very small range across all treatments (83.3-90.6%, with 86.6% in the flooded control) apart from a lower value of 77% in CF-40-CF. There was also a consistent trend for lower straw yield in all stress treatments (except for 10 kPa during FL-PM), but with no significant differences. There was a strong trend for straw yield to decrease as the stress increased in the treatments stressed during FL-PM or at all stages, but with no significant differences. Harvest index (HI) of all treatments with a 40 kPa threshold at any or all stages was significantly lower than that of the CF treatment. A 20 kPa stress during 3L-PI also resulted in significantly lower HI than the CF treatment.

The factorial analysis showed no significant interactions between irrigation threshold and crop stage during which the irrigation treatment was applied for grain yield and all yield components except grain weight (Tables 5 and 6). Grain yield decreased significantly as the irrigation threshold increased from 10 to 20 to 40 kPa. This was largely due to fewer florets per panicle, and to a smaller degree to slightly lower floret fertility (not significant) and grain weight. Grain yield was significantly lower when stresses were applied at 3L-PI or PI-FL than at FL-PM, mainly due to the trend (not significant) for fewer florets per panicle with stresses applied prior to FL. There was significant effect of crop stage on yield and floret fertility (%).

The lowest average grain weight (20.1 mg) was found with a water stress of 40 kPa during FL-PM only, with significantly lower grain weight than for all other stress × stage combinations (except for a stress of 20 kPa during PI-FL) (Table 6). Grain weight is influenced by sink size (number of florets.m^{-2}) which is determined prior to grain filling, and by the amount of photosynthate available for filling the grains. The photosynthate comes from photosynthate stored in the leaves hence biomass at flowering is an important determinant of grain weight and yield (Yoshida, 1981), and from photosynthesis during the grain filling period (Yoshida, 1981). Assimilates during the grain filling period are mainly translocated to the storage organs (*Kropff et al.*, 1994). There was no correlation between grain weight and any of the water stress indices during grain filling. A stress of 40 kPa during FL-PM resulted in significantly lower grain weight than stresses of 10 and 20 kPa at the same stage, and than all stresses at

Table 5. Grain yield, yield components, straw yield and HI as affected by irrigation treatment at different growth stages.

Treatment	Pan pot^{-1} (no.)	Floret. pan^{-1} (no.)	Floret fertility (%)	1000 grain wt (g)	Grain yield (g. pot^{-1})	Straw yield (g. pot^{-1})	HI
			Water stress (W, kPa)				
10	20	94.5	87.9	21.5	35.4	49.2	0.41
20	21	82.4	85.9	21.2	29.6	43.4	0.40
40	20	72.0	82.4	20.8	23.8	43.1	0.35
$LSD_{0.05}$	ns	16.3	ns	0.5	4.9	ns	0.04
			Crop stage (S)				
3L - PI	19	82.1	88.0	21.3	27.9	45.8	0.37
PI - FL	21	80.3	82.3	21.1	27.7	43.4	0.39
FL - PM	21	86.5	85.8	21.1	33.2	46.5	0.40
$LSD_{0.05}$	ns	ns	4.5	ns	4.9	ns	ns
W×S							
$LSD_{0.05}$	ns	ns	ns	0.9	ns	ns	ns

*Data were analyzed by factorial design. 3L=3 leaf, PI=panicle initiation, FL=flowering, PM=physiological maturity, CF=continuously flooded.

Table 6. Interaction of water stress and growth stage on 1000 grain weight (g).

Treatment	Water stress (W), kPa		
	10	20	40
Crop stage (S)			
3L - PI	21.4	21.2	21.4
PI - FL	21.5	20.9	21.1
FL - PM	21.7	21.7	20.1
$LSD_{0.05}$ S×W			0.9

Data were analyzed by factorial design. 3L=3 leaf, PI=panicle initiation, FL=flowering, PM=physiological maturity, CF=continuously flooded.

the other two stages. *Castillo et al.* (2006) also found lower grain weight when drought stress was applied during grain filling. However, in the present experiment, when 40 kPa was applied at all three stages, there was no effect on grain weight.

CONCLUSIONS

With frequent drying, there was no yield reduction for a threshold of 10 kPa applied at 3L-PI or FL-PM, but there was a significant yield decline with 10 kPa applied during PI-FL. However, a threshold of 40 kPa reduced yield by 39-47% when applied at a single stage (3L-PI, PI-FL, FL-PM), while a threshold of 20 kPa reduced yield by 19-33% when applied at a single stage. There was a slightly greater yield reduction (36-48%) when stresses of 20 and 40 kPa were applied throughout all the three stages compared with at a single stage. Once the soil starts drying, soil tension increases rapidly from 10 to 40 kPa, as evidenced by only a very small difference in the number of irrigations applied to each stress treatment within each stage.

RECOMMENDATIONS

The degree of soil drying and the number of drying events are important determinants of crop response to soil drying, and this needs to be considered in setting the irrigation threshold for safe AWD (no yield loss). Given the high sensitivity of rice to water deficit stress as the soil dries beyond 10 kPa, and the fact that there are only small additional water savings in increasing the irrigation threshold above 10 kPa, it is better to adopt conservative AWD practices ie using a threshold of 10 kPa during the vegetative and grain filling stages, and to keep the soil close to saturation during PI-FL.

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Cold Injury and Flash Flood Damage in Boro Rice Cultivation in Bangladesh: A Review

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ABSTRACT

Increasing population with decreasing resources and increasing climate vulnerability appeared as the great challenges to sustain food security of Bangladesh. Cold stress adversely affects growth and productivity of rice (*Oryza sativa* L.). Systematic studies have been carried out to improve understanding on rice cold tolerance. Two adverse conditions, such as low temperature stress at reproductive stages and flash flood at maturity affect the Boro rice in the *haor* areas of Bangladesh. Any deviation of these two phenomena is enough to cause disaster in *haor* areas. Here, we summarized different types of cold injury, rice cold injury scenario and cold tolerant rice varieties/genotypes available in different countries. Moreover, we discussed on rice cold tolerant barrier and flash flood risk in Boro rice cultivation at *haor* areas of Bangladesh. Based on the authors' own research and available data, the concept of overcoming cold and flash flood damage was proposed. According to this concept there were distinguished possible ways how to improve cold tolerance and flash flood problem in Boro rice cultivation in Bangladesh.

Key words: Boro season, cold injury, flash flood, critical temperature

INTRODUCTION

Rice (*Oryza sativa* L.) is one of the most important staple crops and extensively cultivated by more than half of the world's farmers (Fairhurst and Dobermann, 2002; Shelton *et al.*, 2002). More than 90% of the worlds' rice is grown in Asia, where almost 60% of the worlds' population lives (Subudhi *et al.*, 2006). Rice is not only a staple food but also one of the most important cereal crops in Bangladesh which plays a significant role in socio-economy of the country. Enhancing rice production is the key to ensure food security. Rice security is synonymous to food security in many rice growing countries like Bangladesh (Brolley, 2015), which is an important issue for determining social and political stability (Nath, 2015). Rice production in Bangladesh has been increased from 10.79 million tons in 1970 to about 34.86 million tons in 2014-15 (AIS, 2016). Now, Bangladesh is entering gradually into the rice export regime (BER, 2015). However, the future challenge is to maintain the current surplus of rice in order to sustain rice security of the country. Bangladesh will need to produce more food grain with its limited resources to feed her huge population in the coming days.

Various abiotic stresses reduce rice yield greatly (Vij and Tyagi, 2007) which challenge the future food security and are huge threat for agriculture (Kumari *et al.*, 2009). Minimizing yield loss is a major concern to cope with the increasing food demands (Mahajan and Tuteja, 2005). Low temperature induced rice yield loss is a worldwide problem (Peyman and Hashem, 2010), but it is a major constraint to rice production in mountainous regions of the tropics and in the temperate rice-growing zones of the world (Xie *et al.*, 2012). Northern districts of Bangladesh are cold prone areas of the country. Boro rice is greatly affected by cold during crop establishment and reproductive stage. Seedling mortality sometimes goes up to 90% especially in northern part of the country. In recent years, more than 2.0 million hectares of rice crop in northern and north-eastern parts of Bangladesh have been affected by severe cold spell causing partial to total loss. In the *haor* areas of Bangladesh, early planted. Boro

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rice has to face cold stress at the reproductive stages (PI to flowering). Spikelet sterility is observed in this early planted Boro rice, if mean temperature in mid-February to mid-March prevails below 20°C for more than 5-6 days at reproductive stage of the plant (Biswas *et al.*, 2011). The farmers of the *haor* areas consider their Boro crop as a chance harvest as they might have to encounter the flash flood every few years at maturity stage (Biswas *et al.*, 2008). Recent loss due to early flash flood in Boro 2017 at the *haor* area is irreparable. We have to find out a way to cope with the situation. The most important thing to mitigate this issue is cultivation of short duration cold tolerant varieties in the *haor* areas. Since inception in 1971, the scientists in Bangladesh Rice Research Institute are giving their efforts to develop cold tolerant rice varieties. A little success was also made possible with the development of a cold tolerant variety BRRI dhan36, but its efficiency is not praiseworthy in present level of cold stress. In this paper, we have focused on rice cold injury status and cold tolerant rice genotypes in different countries, ways to improve cold tolerance of rice plants and present cold management practices. Moreover, we also discuss on risk reduction in Boro rice cultivation in *haor* areas of Bangladesh.

REVIEW

Low temperature injury in rice

Rice is a thermophilous crop due to its origin in tropical and subtropical regions (Basuchaudhury, 2014). Optimum temperature for rice growing is 25 to 35°C for indica and 20 to 33°C for japonica (Yoshida, l98l). It is more sensitive to cold stress than the other cereal crops such as wheat (*Triticum aestivum* L) and barley (*Hordeum vulgare* L). Low temperature can damage the plant during any development stage such as germination, seedling, vegetative, reproductive and maturity (Andaya and Mackill, 2003; Xu *et al.*, 2008; Ye *et al.*, 2009). The most common symptoms of cold temperature damage at the germination stage are delayed and lower percentage of germination (Cruz and Milach, 2004). At the vegetative stage, chilling damage is expressed through yellowing of the leaves, decreased tillering, lower stature, less biomass production and increased growth duration of the plants (Alvarado and Hernaiz, 2007). Rice is highly susceptible to low temperature at reproductive stage (Yoshida, l98l; Cruz *et al.*, 2006). Low temperature at this stage causes abnormalities at anthesis such as cessation of anther development, non-ripening of pollen grains, non-emergence of anthers from spikelets, partial or no anther dehiscence, little or no pollen shedding and failure of pollen to germinate after reaching stigmas (Cruz *et al.*, 2013).

Cold stress also causes many types of phenotypic damage, such as delayed heading, incomplete panicle exertion and degeneration of spikelet (Shimono *et al.*, 2007; Ye *et al.*, 2009). But spikelet sterility is the most common symptom of injury when rice plants experience cold temperature at the reproductive stage (Jacobs and Pearson, 1994; Cruz *et al.*, 2006). Spikelet sterility results from pollen abortion due to cold injury during microsporogenesis, when pollen grains are being formed at the booting stage (Mackill *et al.*, 1996). The level of damage largely depends on the development stage of rice plants and intensity of the cold event (Jacobs and Pearson, 1994). The young microspore stage is the most susceptible stage to chilling injury that causes male sterility (Yoshida, l98l). This stage occurs approximately 10 to 12 days prior to heading of rice (Heenan, 1984). It is reported that spikelet sterility increases with the increase of low temperature duration at the young microspore stage (Heenan, 1984). Other experiments indicate that 14-7 days before heading, commonly referred to as the booting stage, is considered the most sensitive stage to low temperatures followed by heading or flowering (Yoshida, 1981). Cold stress causes catastrophic yield loss by affecting rice plants' biological functions and overall metabolism negatively. Smillie *et al.* (1988) showed that photosynthesis is impaired at low temperature which reduces growth and yield as there is less carbohydrate available for grain production.

Other studies showed that low temperatures cause delayed and incomplete grain maturation (Ye *et al.*, 2009) that affect adversely on grain yield and quality (Jena *et al.*, 2012).

Critical low temperature

Ambient temperature below 20°C is normally considered as the critical low temperature for rice. The critical temperatures vary from one growth stage to another of the plant (Table 1). But it also differs according to variety, duration of critical temperature, growing conditions and diurnal changes (Farrell *et al.*, 2006). Yoshida (1981) showed that rice plants have a lower threshold temperature (10-13°C) for cold damage during the early stages of development (germination and vegetative), while threshold temperature for damage is higher (18-20°C) at the reproductive stage. Rice plant subjecting to below 20°C at about the reduction division stage of the pollen mother cells usually induces a high percentage of spikelet sterility (Yoshida, 1981). Satake (1976) reported that the thresholds air temperature capable of inducing cold damage at the reproductive stage are 20°C for cold-sensitive varieties and 15°C for cold-tolerant varieties. Crop damage through spikelet sterility occurs if the air temperature prevails below critical low temperature for three days at reproductive stage, but damage is severe if it prevails for more than 5-6 days (BRRI, 2017). Temperatures as low as 12°C will not induce sterility if they last for only two days, but it will induce about 100% sterility if they last for six days (Yoshida, 1981). Farrell *et al.* (2006) reported the critical night temperature capable of inducing cold damage ranges from 13 to 15°C. Biswas *et al.* (2011) suggested night temperature of 12-13°C and day temperature of 28-29°C as critical temperature during reproductive stage of rice plant, as these levels reduced 50% of the original yield level. Although low temperature-induced sterility is normally attributed to low night temperatures, high day temperatures appear to alleviate the effects of low night temperatures (Yoshida, 1981).

Rice cold injury scenario in different countries

Rice cold injury at high latitude and altitude areas were well documented in Japan, Korea, China, Nepal, Bhutan, Australia and other countries (Shimono *et al.*, 2007; Jena *et al.*, 2012). Sthapit (1992) reported that around 26% of rice land of Nepal falls within the temperate region [1000-2000 m above sea level (asl)] and chilling injury is a major constraint of rice production here. The degree of yield loss is quite high at 1300 m above sea level showing high degree of sterility and panicle degeneration (Sthapit, 1992). Gautam and Shrestha (2012) reported that two main types of cold damage occur in Nepal namely delayed-type and sterile-type.

Table 1. Response of the rice plant to varying temperatures at different growth stages[a].

Growth stage	Critical temperature[b] (°C)		
	Low	High	Optimum
Germination	10	45	20-35
Seedling emergence and establishment	12-13	35	25-30
Rooting	16	35	25-28
Leaf elongation	7-12	45	31
Tillering	9-16	33	25-31
Initiation of panicle primordia	15	-	-
Panicle differentiation	15-20	38	-
Anthesis	22	35	30-33
Ripening	12-18	30	20-25

[a]Adapted and modified from Yoshida (1981). [b]Refers to daily mean temperature.

The delayed type of cold damage is prevalent at altitudes between 300 and 1300 m above sea level (Sthapit, 1992; Gautam and Shrestha, 2012). The sterile-type cold damage in rice is usually found at 1300 m above sea level. At Lumlee, (1400 m asl) in Nepal mean air temperature falls below 20°C at the beginning of the reproductive stage. Sterile type of cold damage is a serious problem in this type of environment (Sthapit, 1992; Gautam and Shrestha, 2012). At Chhomrong in Nepal, mean air temperature is low throughout the growing season (Sthapit, 1992). In the Jumla Valley of Nepal, mean air temperature above the critical temperature (20°C) remain only for six weeks in a year from the second week of June to mid August (Sthapit, 1992). The sterile-type cold damage causes incomplete fertilization due to non-viable pollen and failure in anthesis and increases in the number of sterile spikelet (Gautam and Shrestha, 2012; Gautam *et al.*, 2013).

At the higher elevations of Bhutan, air temperatures during seedling growth (March-April) and at later ripening stage (October-November) remain around 15°C or below making the rice varieties highly susceptible to cold injury (Ghimiray, 2012). The temperatures begin to drop sharply from September. If transplanting is delayed, the crop encounters a cold injury at flowering and ripening stages during October-November that cause spikelet sterility (Ghimiray *et al.*, 2013). In India, cold rice is grown in about one million ha of hill regions in Jammu and Kashmir and north eastern hill states accounting 2.3% of total area under rice (Singh *et al.*, 2017). Cold is also an important abiotic constraint at Telangana in India, where low temperature of 8°C to 13°C prevails from December to the first fortnight of February (Neelima *et al.*, 2015).

Low temperatures seriously damaged the Korean rice crop (Lee, 2001). In the northern mountainous regions, the rice plants suffer from low temperatures at any stage between germination and maturity. In years of extreme low temperatures, all rice-growing areas are susceptible to cold at the reproductive stage (Lee, 2001). Early rice suffered from chilling damage mainly at seedling stage (February-March) in southeastern China, while late rice at reproductive stage (October-November) in northeastern, eastern and southwestern China (Dingyuan and Liqun, 1989; Tao *et al.*, 2012). In Japan, air temperature gradually increases from planting of rice (April–May) and reached to the highest peak in August, then it declines to below critical temperature in September and October when rice crops are at flowering or ripening stages. Rice cold damage is a nationwide problem in Japan, but it is a serious problem in the northern regions of Hokkaido and Tohoku areas (Nakagomi, 2013). In Australia, low temperature during late January to early February at reproductive stage disturbs development of pollen grains that causes spikelet sterility (Satake *et al.*, 1987; Reinke *et al.*, 2012).

Variety released worldwide

Many species of tropical or subtropical origin are affected by cold temperatures and exhibit various symptoms of chilling injury such as chlorosis, necrosis or growth retardation. In contrast, chilling-tolerant species are able to grow at such low temperatures (Sanghera *et al.*, 2011). Various rice germplasm accessions exhibit considerable variation in the extent of cold tolerance (Kim and Tai, 2011; Sharifi, 2010). The high mountains of Nepal are important for cold tolerant genes of rice. The famous Jumli Marsi variety of rice is being grown at the highest elevation, up to 3000 meters above sea level (asl) in Chhumchaur, Jumla of Nepal. Gautam and Shrestha, (2012) reported that Chhomrong dhan (rice), Machhapuchre-3 and Palung-2 have been released for cool temperate high hills areas (1,300-2,000 m asl) of Nepal; while Chandannath-1 and Chandannath-3 for Jumla valley (2,300 m asl) and similar areas. On the other hand, Tainan-1, Chainan-2, Chainung-242, Taichung-176, Himali, Kanchan, Khumal-2, Khumal-3, Khumal-4, Khumal-5, Khumal-6, Khumal-7, Khumal-8, Khumal-9 and Manjushree 2 have been released for foot hills to mid hills of warm temperate areas of Nepal. Karki *et al.* (2010) reported that Chhomrong dhan, Machhapuchre-3, Chandannath-1 and

Chandannath-3 and Palung-2 have been adopted so far by farmers in Nepal as chilling tolerant cultivars. Chomrong rice of Nepal has become the most important variety in Bhutan and Madagascar covering around 85% of their rice area.

Barkat a cold tolerant variety in rice-rice rotation in mid altitude valleys of Bhutan was reported as early maturing variety (Ghimiray and Gurung, 1993). Endo *et al.* (2016) reported that a Bhutanese rice variety 'Kuchum' can be useful for developing a cold-tolerant variety. Some other cold tolerant varieties of Bhutan are Jakar Rey Naab, Khangma Maap, Yusi Rey Kaap1, Yusi Rey Kaap2, Yusi Rey Maap1, Yusi Rey Maap2 (Ghimiray *et al.*, 2013). Neelima *et al.* (2015) reported that the rice genotypes Akshaydhan, Taramati, Bhadrakali, RNR 18805, RNR 17813 and WGL 44 were significantly superior to cold tolerant varieties Sheetal, Tellahamsa and JGL 3844 at Telangana in India. Some popular rice varieties grown in high elevation of India and Nepal are HPU1, Kanchan, Himali, Himdhan, Himalaya1, K332, Kalimpong1, Khonorullo and Meghalaya1 (Basuchaudhuri, 2014).

Basuchaudhuri (2014) reported that some varieties from China (B55, Banjiemang, Lijiangheigu and Yunlu 29), Japan (Jyoudeki), the USA (M103 and M104), Hungary (HSC55) and Australia (Quest) were tolerant or moderately tolerant to low temperature at booting and flowering stages. Three varieties from China (B55, Banjiemang and Lijiangheig) and one from Hungary (HSC55) showed consistent tolerance to low temperature at all growth stages. A temperate japonica variety TR22183, developed in northern China, is highly tolerant to low temperatures (Jiang *et al.*, 2011). Some prominent cold tolerant rice varieties in Tohoku region of Japan are Ouu 415, Hitomebore, Ouu-PL 5, Iwate 100, Tohoku 207 and Tohoku PL 3 (Nakagomi, 2013). Sherpa a cold tolerant rice variety had higher spikelet fertility than the commercial cultivars Millin, Quest and Opus in a series of cold-tolerance nurseries, and had spikelet fertility similar to that of the cold-tolerant standard varieties Baijieming and Jyoudeki (Reinke and Snell, 2011). Wang *et al.* (2013) reported that Jinbubyeo, Junganbyeo and SR30084-F8-156 from Korea, IR83222-f8-156 and IR83222-f8-14 from IKO showed cold tolerance with respect to grain fertility.

Rice cold tolerance barrier in Bangladesh

Cold injury of dry season irrigated Boro rice is a common problem for many areas in Bangladesh (BRRI, 2016). During Boro season, air temperature goes below critical temperature of rice especially in the northern part of this country (Figs. 1 and 2). Boro rice experienced very low temperature at seedling stage in December. In some cases, seed germination rate became low. Seedling got yellowish colour with insufficient growth. Farmers were not satisfied with their rice seedlings as they did not get quality seedlings from their seed bed. Seedling mortality occurred after transplanting. Sometimes transplanting was delayed by about two weeks. Some farmers have to buy seedling from market with high price (BRRI, 2016). Generally, Boro rice seeds are sown in seedbed in mid November, transplanted in main field in December and harvested from mid April to early May (BRRI, 2017). However, the farmers of *haor* areas (Sylhet, Sunamganj, Habiganj, Moulvibazar, Netrakona, Kishoreganj and Brahmanbaria) have to transplant Boro rice seedlings earlier than usual planting time to utilize early recession of residual flood water and also to avoid flash flood at the maturity (Biswas *et al.*, 2008; Rashid and Yasmeen, 2017). Planting of Boro rice at usual time do not cause cold injury at reproductive stage in these areas, but crop has to encounter flash flood at maturity (Biswas *et al.*, 2008). On the other hand, early transplanted Boro rice has every probability to face low temperature during February at reproductive phase that caused higher sterility (Fig. 2). Boro crop in the *haor* areas generally matures by the last week of April. Flash flood usually comes in these areas after 2nd week of April as *Boishakhi Dhall* (Biswas *et al.*, 2008). Most of the farmers harvest their Boro rice by this time. But a flash flood by the 1st week of

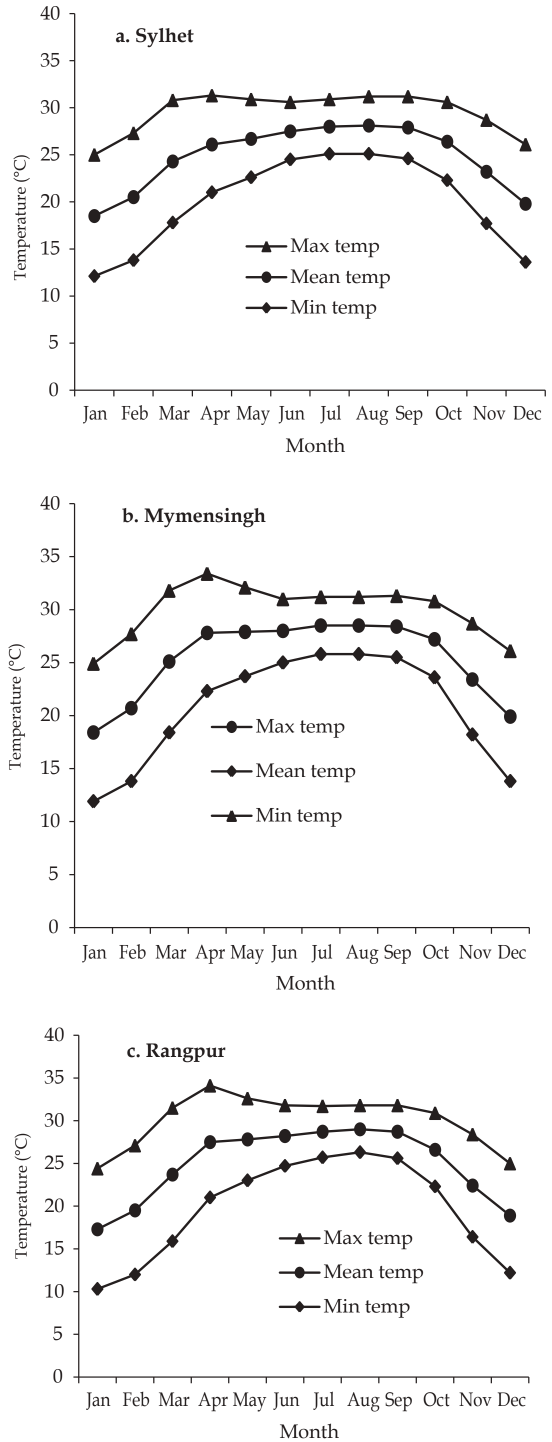

Fig. 1. Month wise maximum, minimum and mean air temperature at a. Sylhet, b. Mymensingh and c. Rangpur, Bangladesh (10-year-average).

April might affect many of the *haor* areas (Rashid and Yasmeen, 2017). Boro rice is submerged at premature stage, if flash flood occurs earlier (ie the 3rd or 4th week of March) as *Chaitali Dhall*, which causes an irreparable damage (Rashid and Yasmeen, 2017). However, early flash flood at premature stage of Boro rice during last week of March to 1st week of April is a rare event. An early flash flood has been devastated in March 2017. Premature Boro rice went under water in many parts of the *haor* areas that caused a huge disaster (Rashid and Yasmeen, 2017).

Analyzing the weather data of Sylhet region it is found that early flash flood was occurred in 1964, 1982, 1996, 2003, 2010 and 2017 from 1956 to till date (Fig. 3). According to Bangladesh Meteorological Department (BMD), the frequency of early flash flood is increasing. The *haor* areas of Bangladesh are sitting just below the downstream of the highest rainfall peak of the world (Cherapunji, Asam) has to face a blow of flash flood every year. Total rainfall for 5-6 days beyond 150 mm at the *haor* basin and its upstream is enough to cause a flood in these areas (Biswas *et al.*, 2008). The rainfall in Sylhet region from 29 March to 3 April had been beyond 150 mm for six times in the years 1964, 1982, 1996, 2003, 2010 and 2017 since 1956 (Fig. 3). It was 156, 191, 153 and 265 mm in 1964, 1982, 2003 and 2010, respectively. In contrast, rainfall was 625 mm in March 2017, the highest ever and 10 times more than normal rainfall (Fig. 3). The rainfall of 2010 and 2017 can be considered as more severe. It has been observed that those abnormal rainfalls were at every seven years. However, the frequency of early flash flood might be closer in the years ahead due to global climate change.

To avoid early flash flood we have to harvest Boro rice by the last week of March. Then a crop will be safe against early flash flood. The growth duration of such variety must be within 130-135 days, if it is seeded and transplanted at usual time. Moreover, it should have some tolerance to cold at reproductive stage (Rashid and Yasmeen, 2017). Variety with such accommodative growth duration is not available in Boro season. Some local

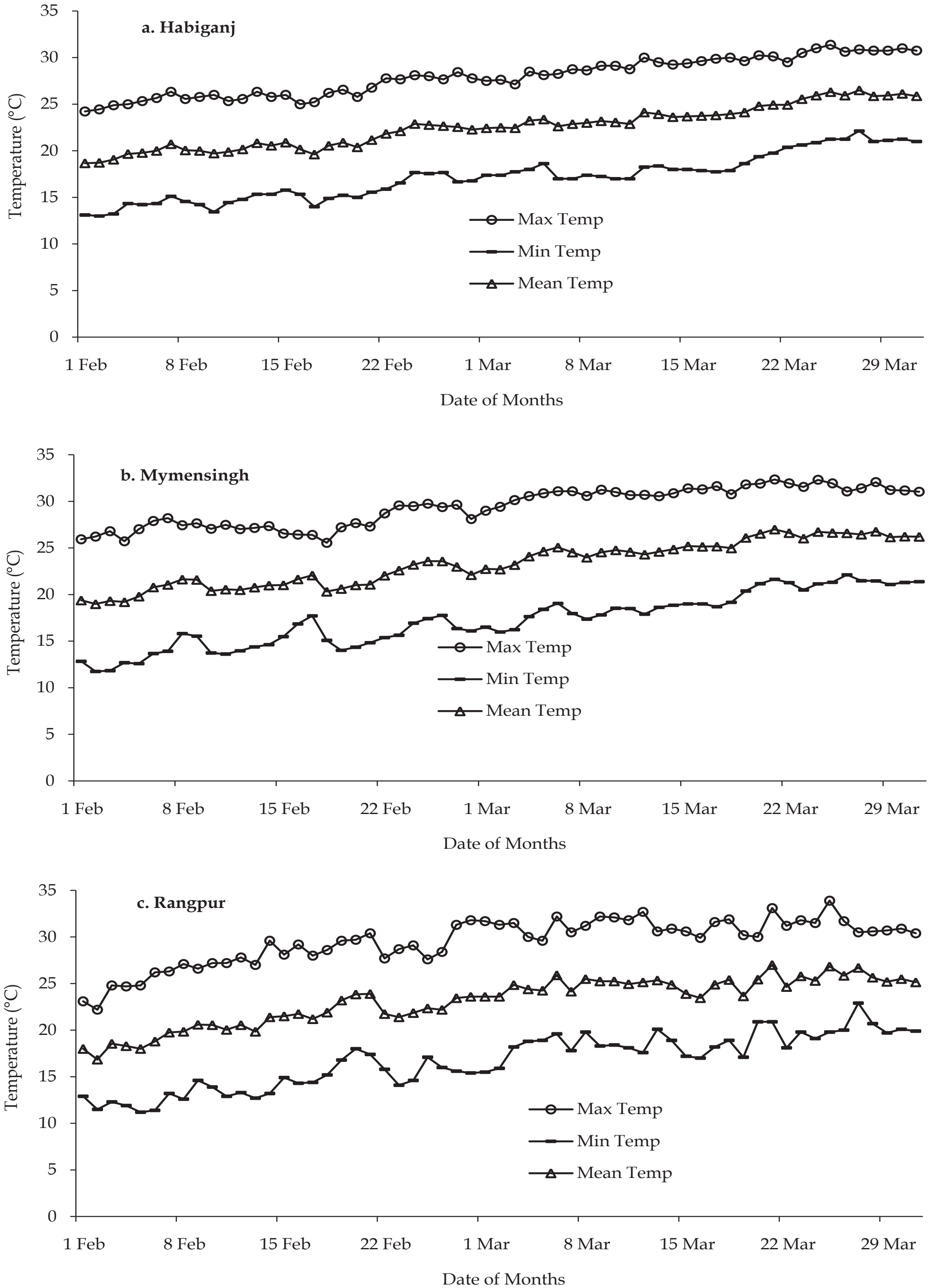

Fig. 2. Air temperature at - a. Habiganj, b. Mymensingh, c. Rangpur during February to March (10-year-average).

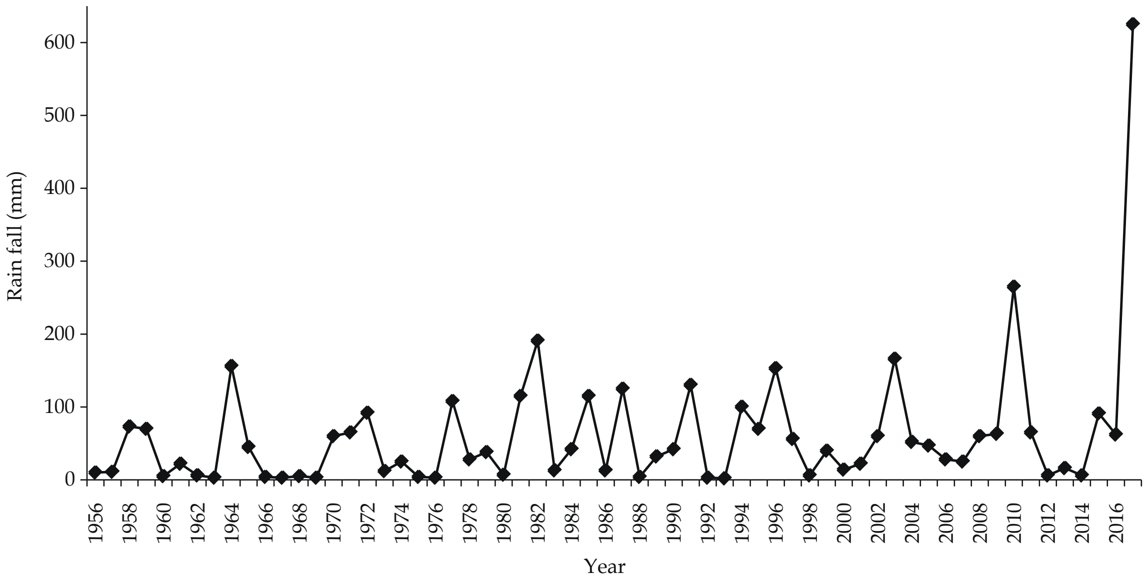

Fig. 3. Total rainfall in Sylhet from 29 March to 3rd April since 1956 to 2017.

improved varieties (LIV) like HbjB-IV and HbjB-VI with 150 days growth duration cannot be accommodated under the stress of early flash flood. The best LIV could yield up to 3.0 t/ha during Boro season. Farmers like to cultivate the best yielding long duration (160 days) variety BRRI dhan29 and they sow seeds in seedbed on first week of November or even last week of October. This crop matures at mid April (Table 2). Generally it did not face cold injury at reproductive stage due to its longer growth duration, but it has a high risk of flash flood at maturity (Table 2). Farmers of the *haor* areas do not prefer to cultivate another short duration Boro variety BRRI dhan28 due its lower grain yield than BRRI dhan29. The growth duration of BRRI developed short duration Boro varieties (BRRI dhan28, BRRI dhan35, BRRI dhan36 and BRRI dhan45) is around 145 days, if recommended cultivation procedure is followed (BRRI, 2017). The appropriate seeding date of a 145-day-old variety (BRRI dhan28) is 15 November (BRRI, 2017). If 30-day-old seedling of this variety is transplanted, crop will be matured by 10 April (Table 2). It means even BRRI dhan28 with the shortest growth duration would not be able to avoid the early flash flood (Table 2).

However, theoretically we can say that any 145-day-variety seeded in 1 November on seedbed would be able to harvest by 25 March that could escape early flash flood. But practically it is not possible. It is observed that when 145-day-variety like BRRI dhan28 or BRRI dhan36 are sown in 1 November and transplanted in first week of December, the crop matures at about 3-4 April (Table 2). Therefore, sowing 15 days earlier than usual time increased the growth duration of these varieties by about a week (Table 2). Moreover, there is a chance of spikelet sterility if short duration variety like BRRI dhan28 is sown in 1 November. So, we should not recommend for seeding of BRRI dhan28 before 15 November. Recently, BRRI has identified moderate level of cold tolerance both at seedling and reproductive stages in a previously developed Boro variety BRRI dhan69. But its growth duration is around one week longer than BRRI dhan28. Sterility problem due to early planting can be minimized by using BRRI dhan69 for its reproductive stage cold tolerance. But, still there will be a risk of the early flash flood (*Chaitali Dhall*) for its longer growth duration (Table 2).

Table 2. Planting time induced sterility and flash flood risk of Boro rice.

Variety	D/S	D/T	PI	Flowering	Harvesting time	Risk
BRRI dhan28, BRRI dhan36	1 Nov	1st week of Dec	1st week Feb (7 Feb)	1st week Mar (4 Mar)	1st week Apr (4 Apr)	Spikelet sterility Early flash flood
	15 Nov	Mid Dec	3rd week Feb (16 Feb)	2nd week Mar (12 Mar)	2nd week Apr (10 Apr)	Early flash flood
BRRI dhan29	1 Nov	Mid Dec	4th week Feb (24 Feb)	3rd week Mar (21 Mar)	3rd week Apr (17 April)	Early flash flood Normal flash flood
	15 Nov	3rd/4th week Dec	1st week Mar (2-3 March)	1st week Apr (2 Apr)	4th week Apr to 1st week May	Early flash flood Normal flash flood
BRRI dhan69	1 Nov	1st week of Dec	3rd week Feb (15 Feb)	2nd week Mar (13 Mar)	2nd week Apr (8 April)	Early flash flood
	15 Nov	Mid Dec	4th week Feb (24 Feb)	3rd week Mar (20 Mar)	3rd week Apr (17 Apr)	Early flash flood Normal flash flood

Mitigation of rice cold injury and flash flood damage

Cultivation of cold tolerant Boro rice varieties is the most appropriate solution to overcome rice cold injury. However, at present we have a few cold tolerant varieties in Bangladesh. BRRI dhan36 has some tolerance to cold but not enough to satisfy farmer. Unfortunately, farmers did not accept this variety. Recently BRRI has identified moderate level of cold tolerance both at seedling and reproductive stages in a previously developed Boro variety BRRI dhan69. Until we have appropriate cold tolerant varieties for Boro season, we have to put emphasis on cultural practices to mitigate cold tolerance. Mahbub *et al.* (2008) reported that Boro seedbed could be covered with transparent polythene sheet at daytime from 10-11 am to sunset for raising healthy seedlings during cold spell. It increases temperature inside polythene cover and ultimately provides better seedling growth. Water management is an important issue for mitigating rice cold injury. Deep watering, round channels and polyethylene tubes around the field can increase water temperature. Use of deep irrigation (20-25 cm) during the reproductive period might help to insulate the crop against low-minimum temperature events (Reinke and Snell, 2011). When the air temperature is low enough to damage rice crops at panicle initiation or early booting, deepwater irrigation (15-20 cm) is an effective way of protecting panicle formation and increasing filled-grain ratio (NYAES, 1993).

Nutrient content in rice plant is an important issue during cold conditions. At low temperature condition during booting, nitrogen concentration in plant should be low. Risk of yield reduction in a cool weather is greatly enhanced by increased N status of crop (Amano, 1984). Top dressing of N at flag-leaf stage during lower temperatures increases the risk of damage to rice crop. Phosphorus is an important nutrient in mitigation of cold injury (Lee *et al.*, 1987). Low temperature damage is reduced when sufficient phosphate is applied in rice plant (RDA, 1981). Organic matter application can improve physical and chemical properties of soil, which may reduce cold damage by increasing water and fertilizer holding capacities in rice field. Amano (1984) showed that applications of compost improved root health and minimized cool weather damage in Japan through reducing spikelet sterility.

Chemical treatments of chilling-sensitive plants lead to increase chilling tolerance. Foliar application of osmoprotectants (spermine and glycinebetaine) offered significant cold tolerance by reducing spikelet sterility in rice cultivars (Zhao *et al.*, 1992; Naidu and Williams, 2004). Chen *et al*. (2005) reported that seed treatment with gibberellic acid and glycinebetaine improves seedling emergence and seedling vigour of rice under low temperature. Exogenous application of abscisic acid (ABA) induced some levels of freezing tolerance in chilling sensitive rice seedlings (Shinkawa *et al*., 2013). Cellular and genetic engineering is a new trend, which allows fundamental changes in the chilling resistance of chilling-sensitive plants. GMO rice plant with trehalose producing genes improves tolerance to drought, salt and low temperature (Wu and Garg, 2003).

For escaping Boro rice crop from flash flood submergence at the *haor* areas of Bangladesh we have to develop Boro rice varieties having growth duration of 130-135 days with reproductive stage cold tolerant. Then rice crop might be safe against early flash flood through earlier harvesting. However, there are some alternatives by which we can harvest Boro rice earlier in the *haor* areas. Double transplanting would be a better option for harvesting one week earlier. In this method of cultivation, rice seeds are to be sown in seedbed on 1 November followed by first transplanting with closer spacing (10 cm ×10 cm) in first week of December at upper parts of the land and finally double transplanting with usual spacing (20 cm × 20 cm) in mid January at lower basins. Manipulation of seedling age could be an alternative option to reduce growth duration. If we transplant 30-day-old seedlings instead of 45-day-old seedlings of same seedbed, the growth duration will be reduced by around a week (Biswas *et al*., 2008). Another alternative could be growing of seedling under polythene shade.

Higher temperature at seedling stage under the shade might compensate some degree-day to reduce its growth duration to some extent. Direct seeding is a way to reduce growth duration by about one week. Sowing sprouted seeds in puddle or zero tillage fields would be a better option. In this method, harvesting could be earlier, but the field duration would be increased. The applicability of all these practices largely depends on the farmer's preference. More importantly, the farmers of the *haor* areas should cultivate short duration Boro varieties (BRRI dhan28, BRRI dhan36, BRRI dhan45, BRRI dhan74, BRRI hybrid dhan3 and BRRI hybrid dhan5) rather than only long duration variety. They have to sow seeds on 15 November and transplant 30-35-day-old seedlings in the main field.

CONCLUSIONS

Cold stress is one of the major abiotic stresses reducing rice production especially in regions where the indica subspecies is cultivated. In Bangladesh, Boro rice seedlings are damaged at seedbed and also after transplanting in December to January, while early planted Boro rice is affected by cold injury at reproductive stage in February. The use of cold tolerant varieties and implementation of appropriate cultural practices are important for minimizing low temperature damage. Moreover, we have to establish the optimum seeding and transplanting time of rice based on minimum, mean and maximum temperatures at different regions with appropriate rice varieties to minimize rice cold injury. Boro rice varieties with shorter growth duration (130 to 135 days), higher yield and tolerant to low temperature at reproductive stage with some desirable traits to the farmer are earnestly needed in the *haor* areas of Bangladesh. Some cold tolerant genotypes are available in different countries. We could collect and use those genotypes in our varietal development programme. Until we develop desired cold tolerant short duration variety, we could try some of the cultural practices able to shorten the growth duration for harvesting earlier in *haor* areas.

- Cultivation of existing short duration high yielding varieties (BRRI dhan28, BRRI dhan36, BRRI dhan45, BRRI dhan81, BRRI dhan84, BRRI hybrid dhan3 and BRRI hybrid dhan5) instead of long duration variety BRRI dhan29.
- Sowing seeds of short duration Boro varieties on 15 November and transplanting 30-35- day-old seedlings in main field.

- Seedling raising under polythene shade to compensate degree day for reducing growth duration.
- Early harvesting (around a week) by using direct seeded rice or manipulation of seedling age or double transplanting method of cultivation.

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Genetic Diversity and Screening of Rice (*Oryza sativa* L.) Genotypes for Drought Tolerance at Reproductive Phase

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ABSTRACT

Drought is a major abiotic constraint for growing rain-fed rice in Bangladesh. A total of 175 rice genotypes were evaluated using field-managed screening protocol to identify reproductive phase drought tolerant genotypes at Bangladesh Rice Research Institute, Gazipur, Bangladesh. Twelve morpho-physiological characters viz, growth duration, plant height, tiller number per plant, panicle number per plant, panicle length, filled grain number per plant, sterility percentage, filled grain weight per plant, 1000 grain weight (TGW), straw weight per plant, harvest index (HI) and percent yield reduction were recorded. Multivariate analysis was carried out by using software Genstat 5.5 to measure genetic divergence among the rice genotypes. In total 175 genotypes were constellated into ten clusters in which the cluster I exhibited maximum genetic distances from cluster V and the lowest genetic distance was between cluster II and X. Inter-cluster distances were higher than the intra-cluster distances suggesting wider genetic diversity among the genotypes of different clusters and homogeneity among the genotypes within the cluster. Among the 12 studied characters percent yield reduction contributed maximum towards total divergence in the genotypes, which revealed that these parameters contributed more to grain yield under drought stress. The genotypes of cluster I namely Canthi bakla (BRRI Genebank Acc. No. 7279), Nizersail (BRRI Genebank Acc. No. 7281), Hashim (BRRI Genebank Acc. No. 7283), Uricheora (BRRI Genebank Acc. No. 7311), Goura Kajol (BRRI Genebank Acc. No. 7312), Chini Sail (BRRI Genebank Acc. No. 7343), Tall Biruin (BRRI Genebank Acc. No. 7355), Sakkar Khora (BRRI Genebank Acc. No. 7506) and Boaincha Biruim (BRRI Genebank Acc. No. 7573) performed better under drought stress, which could be used in the crossing programme as donor parent for the development of variety.

Key words: Genetic distance, drought stress, morpho-physiological characters, multi-variate analysis, yield reduction

INTRODUCTION

Drought is a common feature in Bangladesh especially in dry season (Winter and Pre-monsoon), which causes a substantial reduction of rice yield. It occurs mainly for uneven distribution of rainfall and thus, north-western part of the country is treated as drought-prone (Pervin, 2015). Rice is more susceptible to drought than other cereals. Drought can affect rice plant in any growth stage (Yoshida, 1981). However, T. Aman cultivars usually suffer from drought stress at reproductive and /or early ripening phase resulting poor yield (Pervin, 2015). Rice plant is most sensitive to water stress from panicle initiation to heading stage (Yoshida, 1981). Tuong *et al.* (1995) reported that the reproductive stages such as panicle initiation, panicle development, flowering and anthesis, meiotic development of gametes, fertilization and grain filling are sensitive to water stress, which cause spikelet sterility and rice yield loss. A limited water supply at panicle initiation stage causes 100% and 93% spikelet sterility in variety BR11 and BR22 respectively (BRRI, 1991). Reyniers *et al.* (1982) reported that irreversible damage is caused when water deficit occurs during heading and flowering. Most of the high yielding varieties developed so far are not bred specifically for drought situation. Traditional landraces are important reservoirs of many valuable traits (Hanamaratti

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et al., 2008). Generally, landraces are highly adapted to adverse environment and also have varying levels of resistance to biotic and abiotic stresses (Li *et al.*, 2004). Bangladesh is a land of rice and it has a lot of landraces, which may endure drought sufficiently. Study of diverse genotypes of a crop is necessary to assess their performances, which help to develop a new variety suitable for commercial cultivation. Grouping or classification of genotypes based on suitable scale is quite imperative to understand the usable variability existing among them. Selection of suitable genetically diverse parent to develop heterotic combinations can be facilitated by determining genetic divergence among them. But there is a little work on the analysis of genetic divergence of land race genotypes in relation to drought stress in Bangladesh context. So, the following investigation was carried out to find out the genetic divergence of BRRI rice germplasm and evaluate them for identification of drought tolerant donor.

MATERIALS AND METHODS

In total 175 rice germplasm including BRRI dhan56 and BRRI dhan57 were studied at BRRI farm, Gazipur, Bangladesh during T. Aman season 2015. Field-Managed Screening protocol (IRRI, 2008) was followed for drought screening. The experiment was conducted in two sets where the 1st set was grown under well-watered condition as control and the 2nd set under stress condition. Stress was initiated four weeks after transplanting in which field was drained out properly for not allowing any standing water until maturity. The experiment was laid out in Alpha lattice design with two replications. The perch water table depth and soil moisture was measured daily. Data on growth duration, plant height, tiller number per plant, panicle number per plant, panicle length, filled grain number per plant, sterility percentage, filled grain weight per plant, TGW, straw weight per plant and HI were recorded. The percent yield reduction of tested entries was calculated by comparing rice yield of stress plant with control plant.

Statistical analysis

Mean data for each character were subjected to multivariate analysis viz Principle Component Analysis (PCA), Principle Coordinate Analysis, Non-hierarchical Clustering and Canonical Variate Analysis using Genstat 5.5 [Release 4.1 (PC/Windows NT] (Mahalanobis, 1936; Jager *et al.*, 1983; Digby *et al.*, 1989).

RESULT AND DISCUSSION

Rainfall pattern during experimental period

Flowering of the tested genotypes started from 2nd week of October to 3rd week of November. So, before starting flowering ie during booting stage crop did not receive any rain water at least 13 days although 27.8 mm and 5.6 mm rainfall was occurred on 2nd and 3rd week of October respectively (Fig. 1).

Water table depth

The water table depth was below 30 cm from the soil surface at eight days after drainage of water, while it was about 72 cm for 13 days after drainage (Fig. 2). After that it was again decreased due to rainfall. At 30 days after withholding of water there was no water in the PVC pipe ie the water table depth was around 80 cm below the soil surface.

Soil moisture status

The average soil moisture of the experimental plot was 20.2 to 34.8% during booting to flowering stage (Fig. 3). However, the soil moisture was more than 30% only eight days, which was not consecutive. Consequent crop experiences drought stress at reproductive stage.

Morpho-physiological characters

Significant variations were recorded among the genotypes for all the 12 characters. Table 1 presents latent roots (Eigen values) of 12 principal component axes and percentage of total variations accounted for them obtained from the principal component analysis (PCA). The result revealed that the first axis largely accounted for the variation among the genotypes (95.57%)

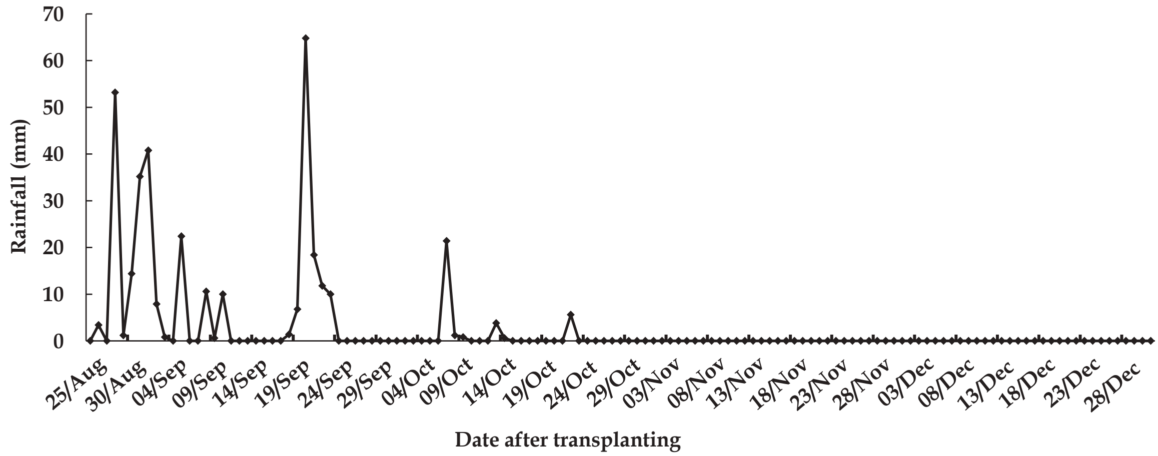

Fig. 1. Daily rainfall at Gazipur during T. Aman 2015.

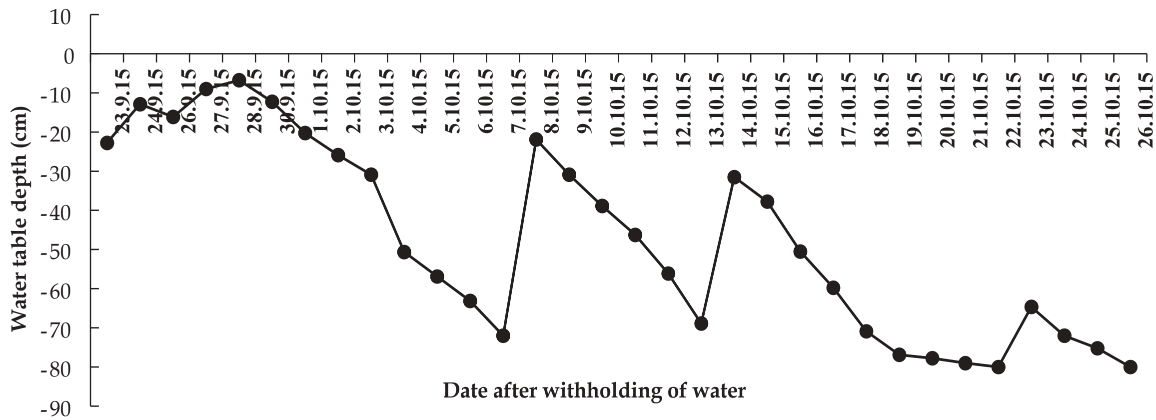

Fig. 2. Water table depth in stress field during T. Aman 2015.

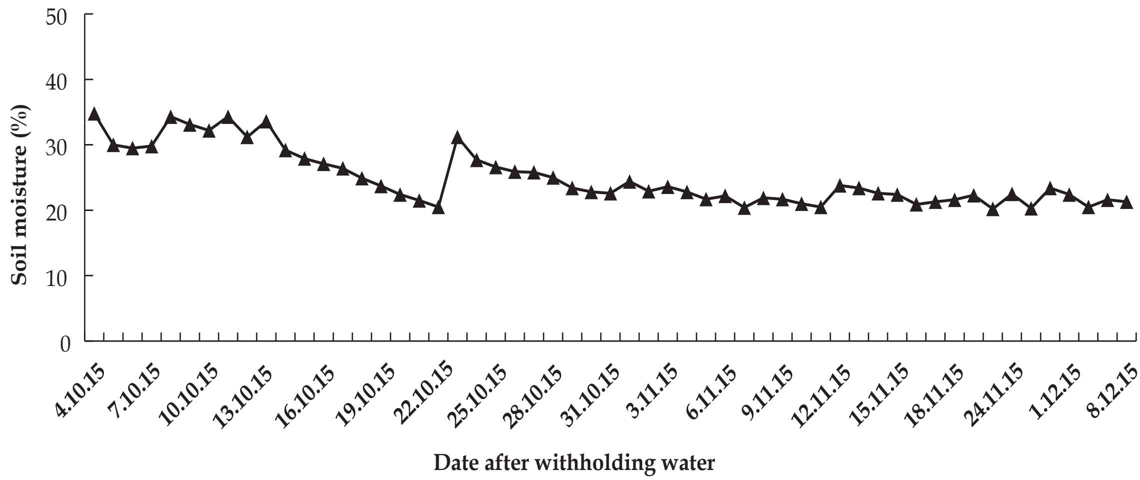

Fig. 3. Soil moisture at experimental plot.

Table 1. Latent roots (Eigen values) and their variations in 12 morpho-physiological characters of 175 rice genotypes.

Principal component axe	Latent root (Eigen value)	Percentage of variation	Cumulative % of variation
1	10927110	95.57	95.57
2	414812	3.63	99.20
3	51080	0.45	99.65
4	17531	0.15	99.80
5	12180	0.11	99.91
6	5561	0.05	99.96
7	3503	0.03	99.99
8	1132	0.01	100.00
9	553	0.00	100.00
10	441	0.00	100.00
11	62	0.00	100.00
12	0	0.00	100.00

followed by the second axes (3.63%). The first two axes accounted about 99% of the total variations among the 12 characters describing 175 genotypes of rice.

The D^2 values ranged from 0.198 to 3.638 and PCA scores also indicated a high degree of genetic diversity among the genotypes (Data not shown). On the basis of D^2 analysis, 175 genotypes of rice were grouped into ten clusters (Table 2). Maximum number of genotypes (25) were included in cluster VII followed by cluster X (24). Cluster V included the minimum number of genotypes (8). Cluster I, II, III, IV, VI, VIII and IX included 9, 20, 15, 17, 23, 14 and 20 genotypes respectively. Table 3 presents the mean values for all the 12 characters along with the marking of the highest (H) and the lowest (L) for each of the cluster. Differences in cluster mean existed for all the characters were studied. In this study, cluster I had the most tolerant genotypes possessing the lowest mean value for percent yield reduction (31.1%) and sterility percentage (33.9%). This group had the highest average in comparison with the other groups considering many traits such as tiller number per plant (14.1), panicle number per plant (12.6), panicle length (24.3 cm), filled grain number per plant (1166.5), filled grain weight per plant (17.9 g), straw weight per plant (31.4 g). Regarding HI, it possesses the second highest value (0.34). Similarly, the genotypes of cluster III showed better performance than the other groups. The percent yield reduction was also the lowest in this cluster (31.2%) and sterility percentage was below 50% (39.6%). Considering traits such as filled grain number per plant (930.8), filled grain weight per plant (16.8 g) and straw weight per plant (28.4 g) this cluster had the second highest average and regarding HI (0.35) this cluster had the highest average among all clusters. The mean values of other characters were also within desirable range in cluster I and III. In contrast to I and III, cluster V had the highest yield reduction (60.0%) and sterility percentage (71.1%). In respect of panicle length (21.4 cm), filled grain number per plant (179.2), filled grain weight per plant (6.7 g) and HI (0.20), it had the lowest mean value. Similar performance was also observed in cluster IX. So the genotypes of cluster V and IX were sensitive to drought stress. Hanamaratti *et al.* (2008) also identified some landraces as good donors of drought tolerance for future breeding programme.

The canonical variate analysis, complementary to Mahalanobis's D^2 statistics, was carried out to obtain the cluster distances (Mahalanobis's D^2 values) that indicated the index of genetic diversity among them. The inter-

Table 2. Distribution of 175 rice genotypes in different clusters through GENSTAT software on the basis of 12 morpho-physiological characters.

Cluster no.	BRRI Genebank Acc. no. of entry	No. of population	Genotype
I	7279, 7281, 7283, 7311, 7312, 7343, 7355, 7506, 7573	9	Canthi Bakla , N. Sail, Hashim, Uricheora, Goura Kajol, Chini Sail, Tall Biruin, Sakkar Khora, Boaincha Biruim
II	7313, 7318, 7349, 7352, 7356, 7497, 7516, 7536, 7564, 7589, 7596, 7604, 7612, 7614, 7615, 7618, 7621, 7637, 7855, 7866	20	Kalokathi Jana, BR11, Ronger Gura, Mohu Madab, Minna Sail, Sada Kumari, Ajal Digha, Kala Biruin, Vaolo, Kaika Biruin (Sada), Khagira, Gabra, Bahori Mota, Monor, Kudi Agroni, Montoysa, Kutiagni Dhan, Kalo Sail, Tepairri, Benapol (Brown).
III	7309, 7314, 7321, 7332, 7344, 7345, 7521, 7547, 7551, 7571, 7583, 7597, 7859, 7864, BRRI dhan56 (ck)	15	Kalo Dhan, Balam, BRRI dhan53, Suganph Dhan, Rash Mala, Chini Sagar, Kali Binni, Bina Sail, Kali Jira (Lal), Deshi Biruir, Khama, Gasta, Kalan Pajam , Hogla Pata, BRRI dhan56 (ck)
IV	7341, 7347, 7357, 7514, 7524, 7555, 7556, 7562, 7575, 7580, 7588, 7599, 7617, 7630, 7643, 7857, 7865	17	BR8922-4-4-4, Malsira, Biruin, Paijam, Biroin, Pak Biruin, Mikal Biruin, Kat Lahi, Neer Dhan, Babusail, Basabo, Gobioha Vhog, Chikon Dhan, Bash Moti, Jamai Kuli, Hasina Chikon, Chini Kanai
V	7327, 7340, 7504, 7511, 7513, 7537, 7613, 7631	8	BR7155-20-1-3, BR6926-1-1-1-3-2, Kakchi Mota, Baro Sail, Akhni Sail, Me-Dhan, Kala Mota, Bambu Dhan
VI	7307, 7316, 7317, IR64, 7328, 7342, 7346, 7503, 7539, 7541, 7550, 7565, 7569, 7572, 7587, 7603, 7611, 7619, 7620, 7628, 7636, 7854, 7861	23	Dakhanalal Dhan, Sakkr Khana, Kala Mota, IR64, Guti Swarna, Tulsi Mala, Kajlo Jira, Kali Satia, Mekli Biruin, Bogla Biruin, Pasu Sail, Bania Chor, Jora Bapail, Chini Gura, Mondol, Tulai Pangi, Bon Hum, Chtrisail, Holud Mota, Mowman Dhan, Ghurum, Shibjata, Nona Khorchi
VII	7277, 7278, 7284, 7285, 7305, 7326, 7333, 7334, 7350, 7354, 7496, 7499, 7505, 7525, 7526, 7549, 7553, 7554, 7560, 7574, 7579, 7600, 7605, 7648, 7863	25	Quchchaly, Nathe Ngepru, NR-1190, Radha, Kalo Aus, BR7770-5 (Nils), Proua-7, Fajla (Nawgan), Chini Kanai, Jhoria, Jol Kumrri, Dudkalam, Kali Jira, Modhu Madhab, Guar Chhora, Bauras, Tri-Dhan, Badsha Bhog, Haitta Binni, Goarchoi, Rosaiya Binni, Dopa, Aman Chala, Boro Bajal, Khejurchori
VIII	7276, 7282, 7320, 7500, 7559, 7566, 7576, 7601, 7602, 7610, 7633, 7646, 7858, BRRI dhan57	14	Ranga Binni, Hashim, BRRI dhan52, Sakkar Khana, Modhu Binni, Lahi, Munsi Biruin, Guti Swarna, Danaguri, Parijat, Lohagura, Hijol Dhiga, Swarna Lata, BRRI dhan57
IX	7300, 7329, 7348, 7353, 7501, 7522, 7523, 7527, 7530, 7535, 7563, 7590, 7608, 7622, 7626, 7634, 7635, 7639, 7642, 7645,	20	Laxmi Digha, Bpt-5204, Nour Sail, Lotma, Sada Mota, Sar Binni, Chenger Muri, Beru Sail, Sada Biruin, Rumu Sail, Kaitta, Kala Biruin, Kajal Hai, Moina Moti, Boleshwas, Holde Mota, Pathar Kuchi, Gaindha, Gachi, Dinga Mony
X	7335, 7351, 7498, 7502, 7507, 7517, 7519, 7529, 7532, 7540, 7545, 7577, 7578, 7585, 7607, 7609, 7616, 7623, 7624, 7632, 7640, 7649, 7852, 7853	24	Kajal (Nawgon), Parabat Jira, Lal Chikon, Lamba Vojon, Khato-Irri, Super Meni, Birol Sail, Lati Sail, Kalo Birun, Nijersail, Swarna Mosori Bhahu Bal, Sini Binni, Satkata Binni, Chanda Binni, Jamli Mota, Buroa Badhe, Khoiya Mota, Kapia Thuti Dhan, Balam, Changai Dhan, Baboni, Jhinuk Mala, Rateil, Munsur

Table 3. Intra-cluster means for 12 morpho-physiological characters in 175 rice genotypes.

Character	Cluster number									
	I	II	III	IV	V	VI	VII	VIII	IX	X
Growth duration (day)	128.4	127.9	126.4	128.2	130.8	131.3 (H)	125.1 (L)	125.2	131.3 (H)	130.8
Plant height (cm)	140.0	135.1	136.1	127.2	115.3 (L)	132.8	160.6 (H)	123.6	121.3	121.3
Tiller no./plant	14.1 (H)	12.3 (L)	13.0	13.6	12.3 (L)	13.1	12.8	12.8	12.6	12.5
Panicle no./plant	12.6 (H)	10.8	11.3	11.7	11.0	11.4	11.0	10.9	10.6 (L)	10.9
Panicle length (cm)	24.3 (H)	22.9	23.2	23.3	21.4 (L)	22.8	23.5	23.0	22.5	22.3
Filled grain (no./ plant)	1166.5 (H)	444.2	930.8	509.7	179.2 (L)	583.1	802.5	689.2	298.5	382.6
% sterility	33.9 (L)	49.5	39.6	51.0	71.1 (H)	43.6	39.8	42.5	60.3	52.6
Filled grain wt (g/plant)	17.9 (H)	11.6	16.8	9.6	6.7 (L)	12.4	14.8	12.7	7.1	8.8
TGW (g)	15.2 (L)	27.7	18.8	22.3	33.5 (H)	23.1	20.1	19.2	23.3	24.9
Straw wt (g/plant)	31.4 (H)	26.8	28.4	23.0	27.1	27.5	25.8	22.6 (L)	24.2	22.7
HI	0.34	0.28	0.35 (H)	0.26	0.20 (L)	0.29	0.33	0.34	0.21	0.26
% yield reduction	31.1 (L)	45.1	31.2	49.9	60.0 (H)	36.6	34.4	36.6	59.5	53.5

Note: H = Highest average value and L = Lowest average value.

cluster distances were higher than the intra-cluster distances in all of the cases reflecting wider diversity among the genotypes of the distant group (Table 4). Similar results were also suggested by Rahman *et al.* (1998) in wheat. The intra-cluster distances were low for all the 10 clusters with the range 0.7918 in cluster IX and 1.2673 in cluster VII that indicated the homogeneous nature of the genotypes within the clusters. Regarding inter-cluster distance, cluster I showed maximum genetic distance (30.42) from cluster V followed by the distance between cluster I and IX (26.82), cluster I and X (24.22), cluster III and V (23.11) and cluster I and II (22.29) suggesting diversity between them and the genotypes in these cluster could be used as parents in hybridization programme. Cluster II had minimum D^2 value (2.23) with cluster X indicating the genotypes in these clusters to be close in genetic make-up. Intermediate or moderate inter-cluster divergence was observed between cluster I and VIII, cluster III and IV, cluster V and VII and cluster VII and IX. Within a certain limit, hybridization between the more diverged parents is expected to generate wide range of variability in segregation generations. Buu and Tuan (1989) also suggested use of diverse genotypes in the hybridization programme for getting transgressive segregants in rice.

Table 5 presents the relative contribution of different characters towards divergence. Vector I and vector II values were obtained from principal component analysis. In first axis vector I, among the 12 studied characters, three characters such as growth duration, % sterility and % yield reduction having positive impact towards divergence. In vector II, four characters such as filled grain weight, TGW,

Table 4. Average inter- and intra- (bold) cluster distance (D^2) for 175 rice genotypes.

Cluster	I	II	III	IV	V	VI	VII	VIII	IX	X
I	**1.2479**									
II	22.29	**0.9553**								
III	7.43	14.97	**1.0677**							
IV	20.31	2.38	13.02	**0.8563**						
V	30.42	8.38	23.11	10.41	**1.1091**					
VI	18.08	4.36	10.76	2.59	12.57	**0.9636**				
VII	11.34	11.05	4.04	9.10	19.28	6.87	**1.2673**			
VIII	14.91	7.59	7.58	5.63	15.81	3.43	3.71	**0.9264**		
IX	26.82	4.78	19.49	6.59	4.27	8.87	15.59	12.08	**0.7918**	
X	24.22	2.23	16.89	4.00	6.56	6.25	12.99	9.46	2.76	**1.0035**

HI and % yield reduction having positive impact towards divergence. The characters that showed positive value in both the vectors contributed most towards divergence. In this study, % yield reduction contributed most for divergence in the studied genotypes than the other characters. The character contributing maximum to the divergence are given greater emphasis for deciding on the cluster for the purpose of further selection and the choice of parents for hybridization (Jagadev *et al.*, 1991). Abd Allah *et al.* (2010) found that number of panicles per plant, TGW, number of filled grains per panicle and panicle weight should be improved in order to increase grain yield under both normal and drought condition.

Table 5. Relative contribution of 12 morpho-physiological characters towards total divergence in rice genotypes.

Character	Vector I	Vector II
Growth duration (day)	0.1708	-0.2818
Plant height (cm)	-0.1113	-0.1915
Tiller no./plant	-0.0502	-0.5361
Panicle no./plant	-0.0670	-0.5348
Panicle length (cm)	-0.1736	-0.0948
Filled grain (no./plant)	-0.4056	-0.1425
% sterility	0.3686	-0.0036
Filled grain wt (g/plant)	-0.4802	0.0136
TGW (g)	-0.0120	0.1131
Straw wt (g/plant)	-0.0361	-0.4264
HI	-0.4312	0.2930
% yield reduction	0.4504	0.0222

From the results of cluster analysis, inter-cluster distance and mean value of studied characters especially yield and yield components under drought stress condition it was observed that cluster I included the most tolerant genotypes and cluster V included the sensitive genotypes. So based on the performance of genotypes under drought stress condition 10 clusters could be classified accordingly (Table 6). Cluster I and III would be ranked as tolerant and obtained score 1, cluster VII and VIII as moderately tolerant and obtained score 3, cluster IV and VI as intermediate and obtained score 5, cluster II and X as moderately sensitive and obtained score 7 and finally cluster V and IX as sensitive and obtained score 9.

CONCLUSION

Based on the results the inter-cluster distances was larger than intra-cluster distances suggesting wider genetic diversity among the

Table 6. Tolerant score and remarks of 175 rice genotypes of 10 clusters.

Cluster no.	Tolerant score	Remark
I, III	1	Tolerant
VII, VIII	3	Moderately tolerant
IV, VI	5	Intermediate
II, X	7	Moderately sensitive
V, IX	9	Sensitive

entries of different clusters but very similar within the cluster. From this study we can concluded that nine genotypes of cluster I namely Canthi bakla (BRRI Genebank Acc. No. 7279), Nizersail (BRRI Genebank Acc. No. 7281), Hashim (BRRI Genebank Acc. No. 7283), Uricheora (BRRI Genebank Acc. No. 7311), Goura Kajol (BRRI Genebank Acc. No. 7312), Chini Sail (BRRI Genebank Acc. No. 7343), Tall Biruin (BRRI Genebank Acc. No. 7355), Sakkar Khora (BRRI Genebank Acc. No. 7506) and Boaincha Biruim (BRRI Genebank Acc. No. 7573) were more tolerant to water stress that could be used as donor parent in hybridization programme.

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Rental Service Market of Farm Machinery in Rice Cultivation: A Farm Level Investigation

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ABSTRACT

This study was undertaken to investigate the rental charge and labour cost of five major operations in rice production in the north-west region of Bangladesh. Farmers' group discussions (FGDs) with 10-15 key informants farmers were conducted for collecting farm level data from 15 villages of both Rajshahi and Rangpur divisions. Rental charge and labour cost of rice cultivation were converted to paddy equivalent cost (PEC) for tillage, transplanting, weeding, harvesting, carrying and threshing. Farmers mostly relied on the service providers of either power tiller or tractor for tillage. They also relied on the service providers of close drum power thresher for threshing rice and other crops. However, farmers still manually carried out transplanting, weeding, harvesting and carrying of rice. Both the contractual and daily basis hired labour and family labour performed those activities. The wages of labour of harvesting and carrying was paid in cash or kind (e.g., share of harvested grain or fixed amount of grain per unit of harvest area). The average paddy equivalent cost for the five major operations was ranged between 37-47% of total paddy production cost in the study locations. The mechanized transplanting and harvesting may reduce rice production cost largely as well as would resolve the labour scarcity problems. Therefore, transplanter and harvester have the great prospect of widespread adoption. However, prospect of mechanical weeding is limited because of the availability of low cost intensive chemical control methods. Thus, providing the larger financial assistance on purchasing both the transplanter and harvester facilitates the end users to enhance the productivity and reduce the cost of rice farming.

Key words: Entrepreneurship, labour, rental charge, paddy equivalent charge, profit

INTRODUCTION

Rice is the main staple food and grown in the three distinct seasons- namely Boro (Dec-April), Aus (April-July), and Aman (Aug-Nov) in Bangladesh. The crop is cultivated over 80% of the total cropped areas in the country (Kabir *et al.,* 2016a). Farmers in some areas of the country compelled to delay transplanting and harvesting rice due to sacrcity of labour at peak period (Sattar, 1999). The scarcity of labour for farming has been increasing due to shifted off-farm wage workers to the non-farm wage work as well as farming is not a preferred livelihood options to younger generation (Kabir *et al.* 2017a; Kabir *et al.* 2017b). Therefore, agricultural mechanization is critically important for sustainable rice production. In this respect, rental system or custom hire service of the farm machinery could be an option to promote farm mechanization in the country (Islam, 2016). In addition, it may create self-employment opportunities for the rural people.

The private entrepreneurs are the main actors of the rental service of farm machinery for land preparation, planting, weeding, and harvesting. They provide services of farm machinery to the farmers on neighbour to neighbour basis.

However, an insufficient support service, poverty and lack of awareness identified as the main constraints of promoting entrepreneurship in Kenya (FAO, 2006). Despite, on average about 51% farmers relied on own machinery and rest dependent on service providers on custom hire basis for tillage and harvesting crops in the southern and central Iraq (Bishay, 2003). Local large-scale landowner provided the rental service with heavy spike-toothed thresher (50% locally manufactured) in Nepal

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(Justice and Biggs, 2010). On the contrary, small-scale enterprises provided the harvesting and threshing services for rice and wheat with the hired combine harvesters to the small and medium size landholders (typically with two acres) in the Gujranwala district of Punjab province in Pakistan. The rental charge varied according to crop density, weed populations, expected yields, ground conditions, and transport distance (Sims *et al.*, 2011).

The farm mechanization in Bangladesh commenced through introduction of irrigation pump for boro rice cultivation with the direction of government in 1970. Thereafter, private entrepreneurship has developed on farm mechanization such as power tractor, pump and thresher. The two wheel and four wheel tractors are locally known as power tiller and tractor respectively in the country. Besides tillage, the power tiller and tractor are widely used for carrying goods with trolley and threshing rice in the off peak tillage season. Most of the farmers including small, medium and some large are relied on the service providers for tillage. Table 1 presents statistics of farm machinery used in rice cultivation in Bangladesh. Few transplanter, weeder, fertilizer applicator, reaper and harvester were used in the country. Mechanized transplanting is being started recently using 4-row walking type transplanter through public and private sector intervention (Islam, 2016). Reaper and harvester is gaining attention to the farmers. Research institutes, department of agricultural extension, private sector and non-government organization are actively involved to promote transplanter, reaper (self propelled or power tiller mounted) and combine harvester in the country.

Transplanting, weeding, harvesting and threshing are the five labour intensive operations in rice cultivation (Islam *et al.* 2016a, Islam *et al.* 2017, Alam *et al*. 2014 and Islam, 2006). Farmers realized the importance of mechanical means of tillage and threshing as to take the enormous benefit of rental system. However, other labour intensive operations such as transplanting, weeding, and harvesting were accomplished manually. The farmers of Bangladesh who are very resource poor often could not afford of buying the machinery– being deprived of the huge benefit of farm power use. There is a need to investigate the existing rental system and labour cost of five labour intensive operations in rice cultivation. Therefore, the present study was undertaken to collect information on the rental system and labour cost of five labour intensive operations to explore the service market of farm machinery in rice production.

Table 1. Present status of farm machinery in Bangladesh.

Machine	Quantity (no.)	Source
Power tiller	7,00,000	Ahmed, 2014
Tractor	60,000	Ahmed, 2014; Kabir, 2014
Rice transplanter	300	Islam, 2016
Weeder	2,50,000	Ahmed, 2014
Granular urea applicator	800	Ahmed, 2014
Prilled urea applicator	18,000	MoA, 2016
Sprayer	13,00,000	Ahmed, 2014
Reaper	500	Ahmed, 2014
Combine harvester	130	Ahmed, 2014; Kabir, 2014
Open drum thresher	1,50,000	MoA, 2016
Closed drum thresher	2,20,000	MoA, 2016

METHODOLOGY

A multistage sampling was applied for this study. At first, 15 districts were selected randomly from the north-west regions of Bangladesh. Thereafter, one upazila was selected randomly from each of the selected districts and a village was selected from each of the selected 15 upazilas. Finally, 10-15 key informant farmers was selected purposively for farmers group discussion (FGD) to delineate with the existing situation of farm machinery rental service. One FGD was conducted in each village to collect data on rental charge of farm machinery, labour cost of major intercultural operations of rice cultivation and farmers opinions about using farm machinery and availability of labour for farming during January-March 2017 (Table 2). The data were summarized in crop production stage wise and discussed in administrative division wise. Labour cost and rental charge of farm machinery in five major operations of rice cultivation were converted to paddy equivalent cost to estimate the percent of total rice production went away for those operations as the rural economy was regulated by the paddy price. The procurement price of boro paddy (Tk 2,400 per ton) was used for estimating paddy equivalent cost of production (DOF, 2017). The average yield of cleaned rice was considered as 3.17 ton per hectare (Kabir *et al.,* 2016b) and used to calculate the rental charge and labour cost of each farming activity in the percentage of paddy production.

RESULTS AND DISCUSSION

Tillage

Farmers in the group discussion said that they used power tractor on rental basis in tillage operation in the case study villages of Rajshahi division. They also mentioned that the service provider only tilled the crop fields; thereafter; the lands owners levelled the land manually for transplanting rice. Table 3 shows that farmers in the survey villages in Chapainawabganj, Naogaon, Natore and Rajshahi districts relied on power tiller for land preparation while farmers in the rest of the locations used both the power tiller and tractor. It was found that irrespective of tractor type, two passes were needed for good pulverization of soil in most of the survey locations except in the Bogra and Rajshahi

Table 2. Study locations.

Division	District	Upazila	Village
Rangpur	Bogra	Sadar	Kanar
	Dinajpur	Sadar	Paschim Khudihar para
	Gaibandha	Gobindaganj	Digholkandi
	Kurigram	Mogolbasha	Uttar Naowabas
	Lalmonirhat	Sadar	Haridev
	Panchagarh	Sadar	10 no. Gorinabari
	Rangpur	Sadar	Ghaghotpara
	Thakurgaon	Sadar	Kismat Daulatpur
Rajshahi	Chapainawabganj	Shibganj	Shekhtola
	Joypurhat	Sadar	Hismi Bazar
	Naogaon	Sadar	Toruk
	Natore	Sadar	Khandarpar
	Pabna	Bera	Fokirkandi
	Rajshahi	Paba	Mohonpur
	Sirajganj	Ullapara	Choria chockpara

Table 3. Rental charge of power tiller and tractor in tilling the land for transplanting rice in Rajshahi division.

District	Power tiller			Tractor		
	Pass no.	Charge Tk *bigha*$^{-1}$	Paddy equivalent charge kg *bigha*$^{-1}$	Pass no.	Charge Tk *bigha*$^{-1}$	Paddy equivalent charge kg *bigha*$^{-1}$
Bogra	4	900	38 (6)	3	1,000	42 (6)
Chapainawabganj	2	700	29 (4)	-	-	-
Joypurhat	2	1,000	42 (6)	2	1,200	50 (8)
Naogaon	2	700	29 (4)	-	-	-
Natore	2	800	33 (5)	-	-	-
Pabna	2	1,000	42 (6)	2	1,000	42 (6)
Rajshahi	4	1,200	50 (8)	-	-	-
Sirajganj	2	1,100	46 (7)	2	1,200	54 (8)
Average	3	938	39 (6)	2	1,100	46 (7)

Source: Field survey 2017. Note: Figures in the bracket is the paddy equivalent charge of tillage as percent of total paddy production in per *bigha* land. One *bigha* equals 33 decimal.

districts. Similarly, it is reported that farmers relied on service providers for tillage in the southern coastal areas in Bangladesh (Kabir *et al*. 2007b). Not surprisingly, per *bigha* rental charge of tractor (Tk 1,000-1,300) was higher than that for power tiller (Tk 700-1,200). It was also the case that per *bigha* paddy equivalent rental charges of power tiller and tractor were 37-50 kg and 41-54 kg, respectively. It can be noted that the paddy equivalent rental charge of tillage either by power tiller or tractor was ranged between 6-7% of total rice production in the per unit area. The key drivers of adoption of four wheel tractors were (i) deep tillage (ii) better pulverization and (iii) faster in tillage operation.

Similarly, in the survey villages in Rangpur division, most of the farmers hired power tiller on rental basis for tilling the lands like that of Rajshahi regions. The key informants mentioned that draft power was used for tillage only in a few plots having difficulties to access power tiller. Farmers said that service providers from other districts came along with their four wheels tractor in Rangpur districts as the demand was very high due to produce fine tilth of soil for sowing potatoes. Table 4 shows that the farmers in the survey villages in Thakurgaon district fully relied on tractor for tilling lands because of the larger size of the plots. On the contrary, farmers in the survey villages in Dinajpur and Panchagarh districts only used power tiller for tillage operation due to smaller size of plots. Similarly, the rental charge of tractor (Tk 900-1100 *bigha*$^{-1}$) in the most areas was higher than that for power tiller (Tk 800-1200 *bigha*$^{-1}$) in the Rangpur division like Rajshahi division, while the number of tillage passes in Rangpur division (3-4 passes) was higher than that of Rajshahi division (2 passes). Similarly, the service providers only tilled the lands in most of the survey villages in Rangpur division like Rajshahi Division, while they tilled and levelled the land for transplanting rice in the Rangpur, Gaibandha and Panchgarh districts. Farmers in Kurigram district expressed that multiple passes were required in tillage operation due to excess weed infestation ultimately increased the cost of tillage. Farmers spent 6-7% of total rice production in the per unit area as paddy equivalent rental charge of tillage.

Transplanting

Farmers transplanted rice manually using family and hired labour in the Rajshahi division. In the group discussion, they mentioned

Table 4. Rental charge of power tiller and tractor in tilling the land for transplanting rice in Rangpur division.

District	Power tiller			Tractor		
	No. of pass	Charge Tk *bigha*$^{-1}$	Paddy equivalent charge kg *bigha*$^{-1}$	No. of pass	Charge Tk *bigha*$^{-1}$	Paddy equivalent charge kg *bigha*$^{-1}$
*Rangpur	3	800	33 (4)	3	1,000	42 (6)
*Gaibandha	4	1,050	44 (7)	3	1,100	46 (7)
Thakurgaon		-	-	3	900	38 (6)
Dinajpur	4	850	35 (5)	-	-	-
Lalmonirhut	3	1,000	42 (6)	3	1,000	41 (6)
Kurigram	4	1,200	50 (8)	4	1,000	41 (6)
*Panchagarh	4	1,100	46 (7)	-	-	-
Average	4	1,000	42 (6)	3	1,000	42 (6)

Source: Field survey 2017, Note: *indicates levelling is complementary with tillage. Figures in the bracket are the paddy equivalent charge of tillage as percent of total paddy production in per *bigha* land. One *bigha* equals 33 decimal.

that the availability of labour in intercultural operations of rice had been decerased in the survey villages. It was due to migration of labourer to urban and cities for non-farm work and educated younger generation preferred non-farm occupations than farming and/or off-farm wage work. Therefore, farmers in the survey villages of Rajshahi, Chapainawabganj, Pabna and Sirajganj districts employed temporarily migrated labourer from other districts for transplanting rice in Boro season. Similarly, farmers in the Natore and Joypurhat districts relied on the migrated labour for transplanting rice in Aman season. In Nagaon district, farmers fully depended on the migrated labourer to transplant rice in both the seasons. Table 5 shows that wage rate and cost of transplanting rice varied among the districts due to availability of wage workers. Labour wages per man-days were varied and ranged between Tk 250-400. It was also the case that the farmers spent paddy equivalent labour wages of transplanting rice ranged between 8-10% of total rice production in per unit area.

Farmers in the group discussion said that both the male and female labourers took part in intercultural operations of agriculture in the survey villages in Rangpur division (Table 6). Key informants noted that there was no scarcity of labourer for intercultural operations of rice as (i) wage work is the main occupation of both the male and female members of landless tribal households and (ii) rate of seasonal labour migration from the areas to cities and other districts was low. The respondents said that per man-day wage of male worker (Tk 300-400) was higher than female worker (Tk 200-250). Rice was transplanted as line or/and traditional haphazard methods in the areas by both contractual and daily wage basis. Table 6 shows that cost of transplanting per *bigha* rice was higher for labourer employed as daily basis (Tk 1,200-1,280) than that for contractual basis (Tk 900-1,000). It can be noted that paddy equivalent cost of labour for transplanting rice in per *bigha* land was ranged between 38-50 kg, which was about 6-8% of total rice production in per *bigha* land.

Weeding

Farmers mainly relied on the human labour for weeding rice fields in the survey villages of Rajshahi division. Table 7 shows that per *bigha* cost of labour for weeding Boro rice among the districts varied largely ie Tk 1,500-2,100 per *bigha* because of the variation in the number of weeding depending on the severity of weed infestation. The paddy equivalent labour cost of weeding per *bigha* land was ranged between 63-88 kg, which was about 10-13% of total paddy

Table 5. Labour cost of manual rice transplanting in Rajshahi division.

District	Labour requirement man-day *bigha*$^{-1}$	Wages Tk man-day^{-1}	Labour cost Tk *bigha*$^{-1}$	Paddy equivalent labour cost kg *bigha*$^{-1}$
Bogra	4	300	1,200	50 (8)
Chapainawabganj	5	250	1,250	52 (8)
Joypurhat	4	400	1,600	67 (10)
Naogaon	4	400	1,600	67 (10)
Natore	5	300	1,500	63 (10)
Pabna	4	300	1,200	50 (8)
Rajshahi	4	400	1,600	67 (10)
Sirajganj	4	400	1,600	67 (10)
Average	4	344	1,444	60 (9)

Source: Field survey 2017, Note: Figures in the bracket are the paddy equivalent labour cost of transplanting as percent of total paddy production in per *bigha* land. One *bigha* equals 33 decimal.

Table 6. Labour cost of manual rice transplanting in Rangpur division.

District	Labour requirement man-day *bigha*$^{-1}$	Wages Tk man-day^{-1}	Labour cost Tk *bigha*$^{-1}$	Paddy equivalent cost of labour kg *bigha*$^{-1}$
Dinajpur	4	320	1,280	53 (8)
Gaibandha	-	Contract	900	38 (6)
Kurigram	4	300	1,200	50 (8)
Lalmonirhat	4	300	1,200	50 (8)
Panchgarh	4	300	1,200	50 (8)
Rangpur	-	Contract	900	38 (6)
Thakurgaon	-	Contract	1,000	42 (6)
Average	4	-	1,097	46 (7)

Source: Field survey 2017, Note: Figures in the bracket is the paddy equivalent labour cost of transplanting as percent of total paddy production in per *bigha* land. One *bigha* equals 33 decimal.

Table 7. Labour cost of manual weeding of Boro rice in Rajshahi division.

District	Labour cost (Tk *bigha*$^{-1}$)			Paddy equivalent labour cost (kg *bigha*$^{-1}$)
	First weeding	Second weeding	Total	
Bogra	1,000	500	1,500	63 (10)
Chapainawabganj	1,100	600	1,700	71 (11)
Joypurhat	1,200	600	1,800	75 (12)
Naogaon	1,100	500	1,600	67 (10)
Natore	1,200	900	2,100	88 (13)
Pabna	1,200	600	1,800	75 (`12)
Rajshahi	1,300	700	2,000	88 (13)
Sirajganj	1,200	800	2,000	83 (13)
Average	1,200	671	1,896	79 (12)

Source: Field survey 2017, Note: Figures in the bracket is the paddy equivalent labour cost of weeding as percent of total paddy production in per *bigha* land. One *bigha* equals 33 decimal.

production in per *bigha* land in the survey villages of Rajshahi division. Normally, two times hand weeding was required to control weed in rice cultivation. However, in some places, additional hand weeding was needed due to severity of weed infestation.

The key informants said that most of the farmers applied one dose of weedicide in the rice fields within 5-7 days after transplanting in the case study villages in the Rangpur division (Table 8). Farmers mentioned that application of herbicides was not only highly cost effective but also not at all harmful for rice crops. Besides, most of the survey villages in Rangpur division, family and hired labourers manually uprooted the weeds from the rice fields once within 25-30 days after transplanting. Per bigha cost of labour for weeding varied largely ie ranged Tk 100–1,500 between the districts in the region because of variation in labour wage (Tk 200-400 per man-day) and severity of weed infestation. On the other hand, per *bigha* weeding cost in Rajshahi division (Tk 1,500-2,100) was largely higher than that for Rangpur division (Tk 1,000–1,500). It was because farmers in the survey villages in Rajshahi division mainly relied on labour for weed control (2-3 manual weeding) while farmers in the Rangpur division employed labour (1-2 manual weeding) as well as applied weedicide (Table 7 and 8). Per *bigha* cost of weedicide was Tk 100-150, which was highly cost effective than manual weeding as application of weedicide saved labour cost for at least one manual weeding (Tk 800-1,200) and reduced labour requirements for following manual weeding because of less weed infestation. The paddy equivalant labour cost of weeding per *bigha* land was ranged between 42-63 kg, which was about 6-9% of total paddy production in per *bigha* land in the survey villages of Rangpur division.

Harvesting

Labour cost of harvesting included harvesting paddy by sickle, binding and carrying to the farmers' homeyard by head and shoulder. Farmers paid cash as labour wage for manual harvesting of rice in most of the survey villages under Rajshahi division except in Naogaon district where about 15% of total harvested paddy was paid to labour as wage (Table 9). Per *bigha* cost of labour for harvesting rice was largely varied (Tk 1,500-2,100) obviously due to variation in the wages between the districts and the distance between crop field to the farmers' home as carrying cost was included in harvesting. The paddy eqivalant labour cost of harvesting per *bigha* land was 67-98 kg, which was about 10-15% of total paddy production in the survey villages of Rajshahi division.

Cropping intensity was high in Rangpur division and labour cost was relatively lower than those of other divisions. Labour scarcity existed in Gaibandha and Kurigram districts

Table 8. Labour cost in manual weeding of Boro rice in Rangpur division.

District	Number of weeding	Labour man-days $bigha^{-1}$	Wages Tk man-day^{-1}	Labour cost Tk $bigha^{-1}$	Paddy equivalent labour cost kg $bigha^{-1}$
Dinajpur	1	4	300	1,200	50 (8)
Gaibandha	1	4	250	1,000	42 (6)
Kurigram	2	6	250	1,500	63 (10)
Lalmonirhat	1	5	200	1,000	42 (6)
Panchgarh	1	3	400	1,200	50 (8)
Rangpur	2	7	200	1,400	67 (10)
Thakurgaon	1	4	300	1,200	58 (9)
Average	1	5	271	1,214	51 (8)

Source: Field survey 2017, Note: Figures in the bracket is the paddy equivalent labour cost of weeding as percent of total paddy production in per *bigha* land. One *bigha* equals 33 decimal.

Table 9. Labour cost of manual harvesting of rice in Rajshahi division.

District	Labour cost Tk *bigha*$^{-1}$	Paddy equivalent cost, kg *bigha*$^{-1}$
Bogra	2,100	88 (13)
Chapainawabganj	1,500	63 (10)
Joypurhat	2,000	83 (13)
Naogaon	6 kg paddy per 40 kg	98 (15)
Natore	1,800	75 (12)
Pabna	2,000	83 (13)
Rajshahi	1,600	67 (10)
Sirajganj	2,100	88 (13)
Average	1,871	80 (12)

Source: Field survey 2017, Note: Figures in the bracket is the paddy equivalant labour cost of harvesting rice as percent of total paddy production in per *bigha* land. One *bigha* equals 33 decimal.

due to migration of labour to the capital. Harvesting cost was observed Tk 2,000 per *bigha* (83 kg paddy) in Kurigram whereas Tk 2,400 (100 kg paddy) in Rangpur district (Table 10). Harvesting cost was varied with season and weather condition. Labour crisis was observed as acute in harvesting boro rice and wage rate increased to Tk 100-200 per man-day from the normal rate due to peak demand. It was estimated that paddy equivalent charge in manual harvesting was 13% (82 kg) of total paddy production in per *bigha* land in the survey areas of Rajshahi division.

Table 10. Labour cost in manual harvesting of rice in Rangpur division.

District	Labour charge Tk *bigha*$^{-1}$	Paddy equivalent cost kg *bigha*$^{-1}$
Dinajpur	2,100	88 (13)
Gaibandha	1,900	79 (12)
Kurigram	2,000	83 (13)
Lalmonirhat	2,000	83 (13)
Panchgarh	1,500	63 (10)
Rangpur	2,400	100 (15)
Thakurgaon	1,800	75 (12)
Average	1,957	82 (13)

Source: Field survey 2017, Note: Figures in the bracket is the paddy equivalant labour cost of harvesting rice as percent of total paddy production in a per *bigha* land. One *bigha* equals 33 decimal.

Reaper

Farmers stated that service providers accepted both the cash and kind (a certain amount of paddy for harvesting per unit area) as rental charge for harvesting rice. Power reaper was used in Bogra and Naogaon districts in the north-west part of Bangladesh. The rental changes of reaper in harvesting rice ranged between Tk 500-550 per *bigha,* which were three to four times lower than that for manual harvesting of rice (Tables 9 and 10). It was noted that carrying cost was not included in the rental charge of harvesting by reaper. Farmers expressed the potential of harvesting equipment in harvesting rice.

Thresher

In the group discussion meeting, farmers mentioned that power operated open drum thresher was widely used for threshing crops in most of the survey villages in the Rajshahi division except Sirajganj district (Table 11). The key informants mentioned that the service providers of threshers accepted both the cash and kind (a certain amount of paddy for threshing 40 kg rice or per unit area) payments for threshing crops. Similarly, they reported that farmers relied on service providers for threshing rice in the southern coastal and north western areas in Bangladesh (Kabir et al. 2007a; Kabir et al. 2007b). On an average, the rental charge of threshers varied from 2.5-3 kg for threshing 40 kg paddy or Tk 600 for threshing

Table 11. Rental charge of thresher in Rajshahi division.

District	Rental charge paddy kg 40 kg^{-1}	Rental charge Tk *bigha*$^{-1}$	Thresher types
Bogra	-	600	ODT
Chapainawabganj	3 (8)	-	ODT
Joypurhat	2.5 (6)	-	ODT
Naogaon	-	600	ODT
Natore	2.5 (6)	-	20% ODT, 80% Pedal thresher
Pabna	3 (8)	-	ODT
Rajshahi	3 (8)	-	ODT
Sirajganj	3 (8)	-	20% ODT, 80% Pedal thresher
Average	2.8 (7)		

Source: Field survey 2017, Note: Figures in the bracket is the paddy equivalant charge of threshing as percent of total paddy production in per *bigha* land. One *bigha* equals 33 decimal. ODT = Open drum thresher.

rice grown in per *bigha* land. It can be noted that cost of fuel and wages of operator was included with the rental charge. Farmers spent 6-8% paddy as rental charge of thresher machine in the surveyed villages of Rajshahi division.

Both the open and close drum threshers were widely used for threshing rice and other crops in Rangpur division (Table 12). Open drum thresher was more preferable than close drum thresher in this region to keep straw long after threshing. Pedal thresher was rarely used in the survey area. Key informants mentioned that a few small and marginal farmers thresh rice by hand beating to save cost in Rangpur district. Farmers replied that a large number of service providers came along with their close drum thresher in Thakurgaon and Panchgarh district from Bogra district to thresh rice and wheat. A group of wage workers and service providers of thresher jointly harvest by sickle and thresh wheat by close drum thresher on a contractual basis in those districts. The rental charge of threshing rice varied from Tk 500-700 among the districts based on the type of threshers. It was estimated that paddy equivalent charge of threshing rice was 3-4% (21-29 kg) paddy in the survey areas of Rangpur division. However, rental charge of thresher machine for whole day operation was Tk 200-250 for pedal and close drum thresher. The farmers paid the cost of fuel and labourers for threshing rice.

On average, farmers paid 37-47% paddy as the service/labour charge of tillage, transplanting, weeding, harvesting and threshing. Labour crisis was observed in transplanting, weeding, harvesting and threshing operation. Labour charge varied widely in transplanting, weeding and harvesting operation. Wide variation of labour charge was observed in harvesting operation due to peak demand and unavailability of labour. Harvesting cost also depended on the distance between crop field to homeyard as carrying cost was included in the harvesting activity. The mechanical intervention in harvesting operation ensured the timeliness and faster operation and reduced the cost of harvesting crops. However, the payback period of transplanter was very high due to seasonal use (Islam *et al.,* 2016 and Islam, 2017). On the other hand, the payback period of reaper was low compared to transplanter and combine harvester (Alam *et al.,* 2014). From the techno-economic point of view, harvesting by reaper may be one solution to reduce the cost of rice production. An entrepreneurship has been developed in the survey villages based on the rental system of farm machinery as tillage and threshing are

Table 12. Rental charge of thresher in Rangpur division.

District	Rental charge Tk *bigha*$^{-1}$	Paddy equivalent rental charge kg *bigha*$^{-1}$	Type of thresher	Remark
Dinajpur	700	29 (3)	CDT	Contract
Gaibandha	500	21 (3)	ODT	Contract
Kurigram	200*	8(1)	Pedal thresher	Machine charge
Lalmonirhat	250*	10 (2)	ODT	Machine charge
Panchgarh	600	25 (4)	ODT	Contract
Rangpur	500	21(3)	ODT	Contract
Thakurgaon	500	21(3)	CDT	Contract
Average	560	23 (4)		

Source: Field survey 2017, Note: Figures in the bracket is the paddy equivalent charge of threshing as percent of total paddy production in per *bigha* land. One *bigha* equals 33 decimal. *Only machine rental charge (excluding labour cost) for whole day operation. CDT = Close drum thresher, ODT = Open drum thresher.

fully mechanized. The entrepreneurs earned money by renting out the close drum thresher for threshing rice and non-rice crops. Labour group purchased thresher and operate it on rental basis. Entrepreneur can invest money in harvester and thresher and operated in combined way to reduce hassle. Large scale financial assistance on the purchase of high value machines like transplanter and harvester may enahnce the mechanized cultivation, increase production and reduce cost.

CONCLUSION

Entrepreneurship of farm machinery in particular power tiller, tractor and thresher were developed and provided service to the farmers across the north-west region of the country. Most of the farmers relied on power tiller and tractor in tillage operation and power thresher in threshing crops. However, there is obvious scarcity of farm machinery in transplanting, weeding and harvesting across the regions. Therefore, farmers relied on family and hired labour in those operations. The rental charge of tillage varied between 6-7% of total paddy production across the locations because of the soil types, number of passes, and tillage types (dry or puddling). The cost of manual transplanting ranged from 7-9% of total paddy production and the weeding cost from 8-12% of total paddy depending on the severity of weed infestation, weeding regime, and application of weedicide. The paddy equivalent wage of harvesting and carrying rice ranged between 12-13% of total paddy production. Finally, the rental charge of thresher ranged between 4-7% of total paddy the farmers produced. The findings indicated that paddy equivalent cost manual of harvesting and carrying was higher followed by manual weeding and transplanting. The cost of weeding might be reduced through increasing use of weedicide. Similarly, the adoption of power reaper or combined harvester may reduce cost of harvesting rice substantially. Thus, the development of entrepreneurship on rice transplanter and harvester has wider scope for self-employment activities.

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Phenotypic Characterization of Jhum Rice (*Oryza sativa* L.) Landraces Collected from Rangamati District in Bangladesh

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ABSTRACT

Genetic diversity of 30 Jhum rice landraces was studied under irrigated condition through Mahalanobis D^2 statistic for agro-morphological characters. The landraces were grouped into five clusters. The inter-cluster distances were higher than intra-cluster distances indicating wider genetic diversity among the landraces of different clusters. The intra-cluster distances were lower in all the cases reflecting homogeneity of the genotypes within the clusters. The principal component analysis exposed that the first five components with vector values > 1 contributed 81.14% of the total variations. The highest number (8) of landraces was observed in cluster IV and the lowest (4) in cluster II. The intra-cluster distance was maximum in cluster III (1.62) containing six landraces and minimum in cluster I (0.57) having six landraces. The highest inter cluster value was 23.44 (between cluster II and V) and the lowest was 7.29 (between cluster III and IV). The highest flag leaf length, leaf area index, filled grains per panicle were found in cluster V while the lowest culm diameter, short duration, low yielding genotypes were clubbed into cluster II. Dwarf stature and lower flag leaf length were recorded into cluster I whereas the highest plant height and panicle length were obtained from cluster IV. Based on canonical vector analysis, culm diameter, days to flowering, days to maturity and length-breadth ratio had maximum contribution towards genetic divergence. Selection of parents from the clusters II and V followed by hybridization would possibly result in desirable transgressive segregants. Finally, Jhum rice landraces need to be conserved in Genebank for future breeding programme.

Key words: Genetic diversity, Jhum rice landraces, agro-morphological characters

INTRODUCTION

Rice (*Oryza sativa* L.) is considered as a major cereal crop in Bangladesh since it is the main staple food for most Bangladeshi people. Rice production area in Bangladesh is about 11.4 million hectares (ha) of land in which 51.64 million tons of rice is produced (BBS, 2015). It constitutes over 91% of the food grain production in Bangladesh. About 76% of the total cropped land is covered by rice and more than 66% of the total agriculture labour force is employed in rice production, processing, marketing and distribution. It provides about 62% of the calorie and 46% of the protein in the average daily diet for the people of the country (HIES, 2010).

In Bangladesh, three major rice crops namely Aus, Aman and Boro constitute 100% of total rice production and grow in three overlapping seasons with large number of varieties that suit various agro-ecological and climatic nitches. The Aus season usually starts in March-April and the crop is harvested in July-August. Aus rices are of two cultural types specifically Broadcast Aus (B. Aus) and Transplanted Aus (T. Aus). B. Aus is direct seeded in dry land rainfed condition. Transplanted Aus is grown under partially irrigated or rainfed condition. Average yield of Aus is 2.00 t/ha in Bangladesh.

The Chittagong Hill Tract (CHT) has an area of 13,180 km, making up approximately 10% of the total area of Bangladesh. Rangamati is the largest district of CHT. Its area is 6116.13

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sq.km and 12 tribes live in Rangamati. The main job of the tribes is mostly crop, livestock, horticulture and forestry agriculture. In ancient days, the tribal people who lived in hilly areas used to practice shifting cultivation, which is locally called Jhum cultivation. It is a unique kind of agricultural practice on sloppy hills of the aboriginal people of CHT region. The method is also known as 'Slash and Burn' or 'Sweden' cultivation. Jhuming compromises cutting and burning of forest trees, clearing spaces and then sowing a variety of seeds. About 20,000 hectares of land are being brought under Jhum cultivation every year. The farmer usually grows low yielding local landraces. More than 300 local Jhum rice landraces have been collected from various locations in CHT, Bangladesh and conserved in Bangladesh Rice Research Institute (BRRI) Genebank (Source: BRRI Genebank accession book). This collection is an invaluable genetic resource that can be used for varietal improvement.

Genetic diversity in the available gene pool is the foundation or the raw material of all plant improvement programme. The availability of transgressive segregants in any breeding programme also depends on effective inclusion of parents. Several genetic diversity studies have been successfully utilized in different crop species based on quantitative and qualitative traits in order to select genetically distant parents for hybridization (Bedoya *et al.*, 2017; Ahmed *et al*., 2016a; Islam *et al*., 2016; Malek *et al*., 2014; Khodadadi *et al*., 2014).

Bangladesh has abundant diversified rice landraces from time immemorial. More than 8,000 rice germplasm have been registered in BRRI Genebank (BRRI, 2016). Among them, about 1,500 Aus rice landraces also have been conserved in BRRI Genebank. But scant information is available on genetic divergence about hilly rice landraces especially in Rangamati district. Keeping this in view, the present study was focused to assess the genetic diversity of 30 Jhum rice landraces using Mahalanobis D^2 statistics.

MATERIALS AND METHODS

Collection of Jhum rice landraces

A total of 30 Jhum rice landraces (Table 1) were collected from Rangamati districts of Bangladesh during 24-28 October, 2015 by Dr M Z Islam and colleagues, Bangladesh Rice Research Institute (BRRI) under Asian Food and Agriculture Cooperation initiative (AFACI) project. All rice landraces were grown under Jhum. Fresh seed stocks for each landrace were collected from fields and farmer's store. The collected landraces have been conserved at short term storage of the BRRI Genebank.

Characterization of germplasm for agro-morphological traits

The collected Jhum rice landraces were characterized for agro-morphological traits. These studies were conducted in RCB design with three replications at the experimental field of BRRI, Gazipur, during March to July (Aus season) 2016. Twenty-day-old seedlings from each entry were transplanted using single seedling per hill in 2.4 m^2 plot. Row to row and plant to plant distances were maintained as 25 cm and 20 cm, respectively. Fertilizers were applied @ 60:20:40:12 kg N, P, K and S per hectare. All the fertilizers except N were applied at final land preparation. Nitrogen was applied in three equal splits, at 15 days after transplanting (DAT), at 35 DAT and just before flowering. Intercultural operations and pest control measures were done as and when necessary.

Twenty-three qualitative traits namely blade pubescence, blade colour, leaf sheath: anthocyanin colour, basal leaf sheath colour, flag leaf angle, ligule colour, ligule shape, coller colour, auricle colour, culm:anthocianin colouration of nodes, culm angle, internode colour, culm strength, panicle type, panicle exertion, spikelet: awns in the spiklet, distribution of awing, awn colour, apiculus colour, stigma colour, lemma and palea colour, seed coat (bran) colour and leaf senescence were scored based on 'Descriptors for cultivated

Table 1. Information on local name, place of collection, season and origin of the Jhum rice landraces.

Name	Upazila	District	Season	Origin
Kobrok-1	Rangamati Sadar	Rangamati	Jhum	Bangladesh
Vanguri Jhum	″	″	″	″
Amey-1	″	″	″	″
Horinbinni	″	″	″	″
Bairi	″	″	″	″
Lonkapara binni	″	″	″	″
Turni	″	″	″	″
Vanguri valo	″	″	″	″
Bandornok binni	″	″	″	″
Kamarang-1	″	″	″	″
Lokkhi binni	″	″	″	″
Gonda	″	″	″	″
Kangbui	″	″	″	″
Koborok-2	″	″	″	″
Galong	″	″	″	″
Chorui	″	″	″	″
Pattiki	″	″	″	″
Amey-2	″	″	″	″
Guri	″	″	″	″
Kamarang-2	″	″	″	″
Suri	″	″	″	″
Kamarang-3	″	″	″	″
Amey-3	″	″	″	″
Badoi	″	″	″	″
Turki	Kaptai	″	″	″
Kangbui	″	″	″	″
Kongcho	″	″	″	″
Bidi	″	″	″	″
Kobrok	″	″	″	″
Mongkhoi	″	″	″	″

rice (*Oryza sativa* L.)' issued by GRSD, BRRI (2018) (Table 2). Again, specifically ten plants from each entry were randomly selected for recording data on 15 quantitative traits namely, flag leaf length, flag leaf width, leaf area index, culm diameter, plant height, effective tiller number, panicle length, days to flowering, days to maturity, filled grains per panicle, grain length, grain breadth, length-breadth ratio, 1000 grain weight (TGW) and yield per hill. Genetic diversity was worked out by the principal component analysis (Rao, 1964) and Mahalanobis' generalized distance (D^2) analysis (Rao, 1952). Intra and inter cluster distances were calculated by the methods of Singh and Chaudhury (1985). Under multivariate analysis principal component analysis (PCA), principal coordinate analysis (PCoA), cluster analysis (CA) and canonical vector analysis (CVA) were done by using GENSTAT 5.5 programme.

RESULTS AND DISCUSSION

Collection and characterization of existing germplasm is not only important for utilizing the appropriate attribute based donors in breeding programmes, but is also essential in the present era for protecting the unique rice. Using advanced biometric techniques such as

Table 2. Classification of Jhum rice landraces based on 23 qualitative characters.

Character	Classification	Total variety	Landraces (Serial number in Table 1)	Frequency %
Blade pubescence	01. Glabrous	01	9	3.33
	02. Intermediate	28	1,3,4,5,6,7,8,10,11,12,13,14,15,15,16,17,18,19,20,21,22,23,24, 25,26,27,28,29,30	93.34
	03. Pubescent	01	2	3.33
Blade colour	01. Pale green	01	22	3.33
	02. Green	28	1,2,3,4,5,7,8,9,10,11,12,13,14,15,16,17,18,19,20,21,23,24,25,2 6,27,28,29,30	93.34
	05. Purple margins	01	6	3.33
Leaf sheath: anthocyanin colour	01. Absent	28	1,2,3,4,5,7,8,9,10,11,13,14,15,16,17,18,19,20,21,22,23,24,25,2 6,27,28,29,30	93.33
	09. Present	02	6,12	6.67
Basal leaf sheath colour	01. Green	28	1,2,3,4,5,7,8,9,10,11,13,14,15,16,17,18,19,20,21,22,23,24,25,2 6,27,28,29,30	93.34
	02. Purple lines	01	6	3.33
	03. Light purple	01	12	3.33
Flag leaf angle	01. Erect(<30^0)	10	1,2,3,8,14,16,17,18,24,27	33.33
	03. Semi erect(<30-45^0)	06	7,11,19,23,26,30	20
	05. Horizontal (<46-90^0)	09	6,9,15,20,21,22,25,26,28	30
	07. Descending (>90^0)	05	4,5,12,13,29	16.67
Ligule colour	01. White	29	1,2,3,4,5,7,8,9,10,11,12,13,14,15,16,17,18,19,20,21,22,23,24,2 5,26,27,28,29,30	96.67
	03. Purple	01	6	3.33
Ligule shape	02. 2- cleft	30	1,2,3,4,5,6,7,8,9,10,11,12,13,14,15,16,17,18,19,20,21,22,23,24, 25,26,27,28,29,30	100
Collar colour	01. Pale green	29	1,2,3,4,5,7,8,10,11,12,13,14,15,16,17,18,19,20,21,22,23,24,25, 26,27,28,29,30	96.67
	02. Green	01	9	3.33
Auricle colour	01. Pale green	30	1,2,3,4,5,6,7,8,9,10,11,12,13,14,15,16,17,18,19,20,21,22,23,24, 25,26,27,28,29,30	100
Culm anthocyanin colour	01. Absent	27	1,2,3,4,5,7,8,9,11,12,13,14,16,17,18,19,20,21,22,23,24,25,26,2 7,28,29,30	90
	09. Present	03	6,10,15	10
Culm angel	01. Erect(<30^0)	07	4,6,12,14,17,24,27	23.33
	03. Intermediate	13	1,3,5,7,9,13,16,20,21,23,25,26,30	43.33
	05. Open	10	2,8,10,11,15,18,19,22,28,29	33.33
Internode colour	01. Green	22	1,3,5,7,8,9,13,14,16,17,19,20,21,22,23,24,25,26,27,28,29,30	73.33
	02. Light gold	05	2,4,11,12,18	16.67
	03. Purple lines	03	6,10,15	10
Culm strength	01. Strong	18	1,2,3,6,7,8,10,12,13,14,15,17,20,21,24,25,27,28	60
	03. Moderately strong	11	5,9,11,16,18,19,22,23,26,29,30	36.67
	05. Intermediate	01	4	3.33
Panicle type	01. Compact	01	11	3.33
	05. Intermediate	06	4,6,15,26,29,30	20
	09. Open	23	1,2,3,5,7,8,9,10,12,13,14,16,17,18,19,20,21,22,23,24,25,27,28	76.67
Panicle exsertion	01. Well exerted	13	1,2,3,4,5,6,7,9,14,16,18,19,21	43.33
	03. Moderately well exerted	09	8,12,15,17,20,23,24,26,28	30
	05. Just exerted	08	10,11,13,22,25,27,29,30	26.67

Table 2. Contined.

Character	Classification	Total variety	Landraces (Serial number in Table 1)	Frequency %
Spikelet: awns in the spikelet	01. Absent	28	1,2,3,4,5,6,7,8,9,10,11,12,13,14,15,16,18,19,20,21,23,24,25,26,27,28,29,30	93.33
	09. present	02	17,22	6.67
Distribution of awning	01. Tip only	02	17,22	6.67
Awn colour	01. Straw	02	17,22	6.67
Apiculus colour	02. Straw	17	3,10,11,13,14,16,17,19,20,21,22,23,24,26,28,29,30	56.67
	05. Red apex	04	1,15,25,27	13.33
	06. Purple	09	2,4,5,6,7,8,9,12,18	30
Stigma colour	01. White	26	1,2,3,4,8,9,10,11,12,13,14,15,16,17,18,19,20,21,22,23,24,25, 26,28,29,30	86.67
	05. Purple	04	5,6,7,27	13.33
Lemma and palea colour	0. Straw	25	1,2,3,5,8,10,11,12,13,14,15,16,17,18,19,20,21,22,23,24,25,26,27,28,30	83.33
	01. Gold	01	29	3.33
	03. Brown furrows on straw	01	6	3.33
	05. Reddish to light purple	02	4,9	6.67
	07. Purple furrows on straw	01	7	3.33
Seed coat (bran) colour	01. White	26	1,2,3,4,7,8,9,10,11,12,13,15,17,18,19,20,21,22,23,24,25,26,27,28,29,30	86.67
	05. Red	03	5,14,16	10
	07. Purple	01	6	3.33
Leaf senescence	01.Late and slow	01	9	3.33
	05. Intermediate	29	1,2,3,4,5,6,7,8,10,11,12,13,14,15,16,17,18,19,20,21,22,23,24,25,25,26,27,28,29,30	96.67

multivariate analysis based on Mahalanobis D^2 statistic (Mahalanobis, 1936), it has now become possible to quantify the degree of genetic divergence amongst populations and assessing the relative contribution of various desirable attributes of breeding and agronomic value to the total divergence by the clustering pattern. The grouping of genotypes into so many clusters had suggested the presence of high degree of diversity in the materials evaluated.

Qualitative traits characterization

Among the investigated 38 agro-morphological characters, 23 characters were qualitative. Qualitative characters are important in respect to the identification and the characterization of landraces of rice varieties because it was observed that these characters are less influenced by the various environmental conditions. Polymorphism was found in 21 of the 23 qualitative traits under studied; the non-polymorphic traits were ligule shape and auricle colour (Table 2). The detailed phenotypic characterizations of Jhum rice landraces for 23 qualitative agro-morphological characters are as follows:

Leaf traits

Jhum rice landraces were characterized for leaf traits at late vegetative and flowering stages. Most of the characterized Jhum rice (28) landraces (93.34%) exhibited intermediate leaf blade while the rest of the landraces showed pubescent (3.33%) and glabrous (3.33%) type. Similarly, it was observed in the present study that 28 landraces had green (93.34%) color leaf blade, while one had pale green (3.33%) and one had purple margin (3.33%) colour leaf blade. However, the anthocyanin colouration of leaf sheath was present only on two landraces (6.67%), while the rest (28) had no coloration (93.33%) of anthocyanin. The maximum number of landraces (33.33%) had erect (<30°) type of flag leaf blade, while (20%) of each had semi-erect (<30-45°) and (30%) horizontal (<46-90°) and only 16.67 % had descending (>90°) type of flag leaf.

Ligule traits

Ligule colour of 29 landraces was observed white (96.67%) type colour while the remaining only one lonkapara binni landrace showed purple colour. There was no divergence found in ligule shape for the studied landraces. All the landraces were observed 2-clefted. Again, most of the landraces (29) showed pale green (96.67%) collar colour while the remaining just one landrace namely bandornok binni was observed having green collar colour (3.33%).

Culm traits

The qualitative characters showing higher variability were culm angle (23.33% erect, 43.33% intermediate and 33.33% open), internode colour (73.33% green, 16.67% light gold and 10% purple line), culm strength (60 % strong, 36.67% moderately strong and 3.33% intermediate).

Panicle traits

On the basis of attitude of panicle, 23 landraces had open type (76.67%) of panicle among the 30 landraces. Six landraces had intermediate type (20%) of panicle and rest of the one landrace namely Lokkhi binni had compact type (3.33%) of panicle. Among the 30 landraces of Jhum had well exerted (43.33%), moderately well exerted (30%) and just exerted (26.67%) on the basis of panicle exersion.

Grain traits

Based on awns in the spikelet, most of the 28 landraces awns were absent (93.33%) and two landraces (Pattiki and Kamarang-3) had present awns (6.67%). Again, two landraces awns were distributed on the tip only and colour was straw. The spikelets showed straw (56.67%), red apex (13.3%) and purple (30%) colour apiculus. On the basis of stigma colour, white (86.67%) and purple (13.33%) colour was observed in 28 and two landraces, respectively. Among the 30 landraces, various colours were observed in husk (lemma and palea) of different landraces. Straw colour (83.33%) lemma and palea were recorded in 25 landraces, gold (3.33%) in one, brown furrows on straw (3.33%) in lonkapara binni and reddish to light purple in two landraces namely Horinbinni and Bandornok binni. Also, one landraces Turni had purple furrow on straw (3.33%). Around, 26 landraces showed white (86.67%) seed coat colour, while three (Bairi, Koborok-2 and Chorui) and one (Lonkapara binni) landrace had red (10%) and purple (3.33%) seed coat colour, respectively. Maximum number of landraces 29 had intermediate type (96.67%) and one landrace had late and slow type (3.33%) of leaf senescence at the time of maturity among the 30 Jhum rice landraces.

The present study exhibits high variability in most of the observed qualitative traits of Jhum rice landraces. Similar findings were reported by Akter *et al.* (2017), Mau *et al.* (2017), Ahmed *et al*. (2016b), Nascimento *et al*. (2011) and Moukoumbi *et al.* (2011). However, Ahmed *et al*. (2015a) characterized 21 genotypes of similar or duplicate named Kartiksail rice germplasm of Bangladesh and showed no anthocyanin colour in auricles, weak and medium intensity of anthocyanin colour in basal leaf sheath (each 5%), green leaf blade colour (47%) and white colour of stigma (90%). Parikh *et al.* (2012) also observed majority of the genotypes to possess green basal leaf sheath colour (84.5%), green leaf blade colour (86.8%), white ligule colour (94.7%), light green auricle colour (97.3%), semi erect plant habit (44.7%), straw apiculus colour (53.9%) and awnless (72.3%) in 71 aromatic rice germplasm.

Quantitative traits characterization

Table 3 presents Eigen values (latent roots) and percentage of total variation accounted for them obtained from principle component analysis. The result exposed that the first five components in the PCA with Eigen values >1.0, contributed 81.14% of the total variations among the genotypes for 15 morphological characters. These results are in agreement with the findings of Ahmed *et al.* (2015b). Fig. 1 presents a two dimension chart (Z_1-Z_2) of the landraces based on principal axes. As per the scattered diagram, the Jhum rice landraces were apparently distributed into five clusters.

The clustering pattern was determined by the principal component analysis (PCA), which represents composition of different clusters with the landraces (Table 4). Maximum 8 landraces

Table 3. Latent roots (eigen values) and their variation in 15 quantitative characters in 36 Jhum rice landraces.

Character	Principal component axes	Latent root	Variation (%)	Cumulative % of variation
Flag leaf length (cm)	PC 1	3.890	25.94	25.94
Flag leaf width (cm)	PC 2	3.179	21.20	47.14
Leaf area index (cm^2)	PC 3	2.350	15.67	62.81
Culm diameter (mm)	PC 4	1.497	9.98	72.79
Plant height (cm)	PC 5	1.253	8.35	81.14
Effective tiller number	PC 6	0.883	5.89	87.03
Panicle length (cm)	PC 7	0.812	5.42	92.45
Days to flowering	PC 8	0.380	2.54	94.99
Days to maturity	PC 9	0.271	1.81	96.8
Filled grains per panicle	PC 10	0.210	1.41	98.21
Grain length (mm)	PC 11	0.156	1.04	99.25
Grain breadth (mm)	PC 12	0.101	0.67	99.92
Length-breath ratio	PC 13	0.006	0.04	99.96
1000 grain weight (g)	PC 14	0.004	0.03	99.99
Yield/hill (g)	PC15	0.001	0.01	100

Table 4. Distribution of 30 Jhum rice landraces into five clusters.

Cluster	No. of landraces	% total	Landrace
I	6	20.00	Amey-1, Horinbinni, Bandornok binni, Kamarang-1, Suri, Kobrok
II	4	13.33	Lonkapara binni, Gonda, Amey-2, Kamarang-2
III	6	20.00	Koborok-2, Galong, Chorui, Kamarang-3, Bidi, Mongkhoi
IV	8	26.67	Kobrok-1, Bairi, Turni, Vanguri valo, Kangbui, Pattiki, Guri, Turki
V	6	20.00	Vanguri Jhum, Lokkhi binni, Amey-3, Badoi, Kangbui, Kongcho

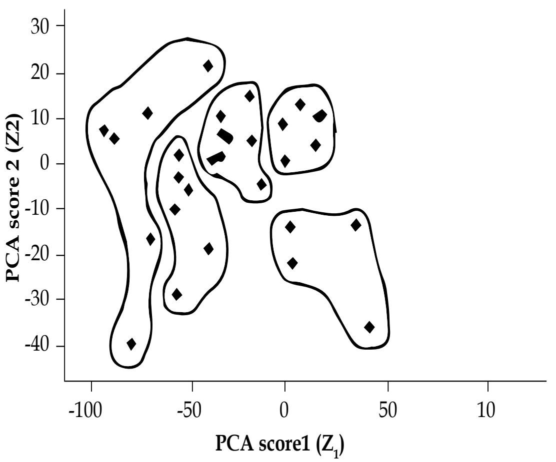

Fig. 1. Scatter diagram of 30 Jhum rice landraces based on their principal component scores superimposed with clustering.

were grouped into the cluster IV followed by six in cluster I, III and V. The cluster II contained the lowest (4) number of landraces. Siddique *et al.* (2011) found five clusters from 38 rice landraces of hilly areas of Bangladesh. Similarly, Khatun *et al*. (2015) studied five clusters from 43 upland rice genotypes. Again, Roy *et al.* (2004) found five clusters from 35 Aman rice cultivars for 10 traits during Kharif season.

Table 5 shows results of five higher and five lower inter genotypic distances estimated from distant matrix of Principal Coordinate Analysis. The highest inter genotypic distance was 1.22 observed between Koborok-2 and Chorui followed by the distance of 1.20 observed between Galong and Bidi. The lowest distance was calculated (0.37) between Bairi

Table 5. Five higher and five lower inter genotypic distance among the 30 Jhum rice landraces.

Genotypic combination	Distance
A. Five higher inter genotypic distance	
Koborok 2- Chorui	1.22
Galong- Bidi	1.20
Kamarang 1-Suri	1.17
Lonkapara binni- Amey 2	1.13
Bandornok binni- Suri	1.11
B. Five lower inter genotypic distance	
Bairi - Kangbui	0.37
Vanguri valo- Turki	0.39
Horinbinni- Bandornok binni	0.43
Bairi- Vanguri valo	0.47
Bandornok binni- Kamarang 1	0.50

and Kangbui followed by the distance of 0.39 observed between Vanguri valo and Turki. Buu and Tuan (1989) and Joshi and Dhawan (1966) reported that genetic diversity is very much important factor for any hybridization programme aiming at genetic improvement of yield especially in self-pollinated crops like rice. Again, Bhatt (1970) inferred that Mahalanobis's D^2 statistic is a powerful tool for choosing diverse parents for hybridization.

Table 6 presents the average intra and inter-cluster distances (D^2). The intra-cluster distance was maximum in cluster III (1.62) containing six genotypes followed by cluster II (0.85) with four genotypes and minimum in cluster I (0.57) having six genotypes and the cluster IV showed the second lowest intra-cluster distance (0.68) having the highest (8) number of genotypes. Such results indicated that the landraces of cluster III were the most diverse and those of cluster I was most similar or less diverse.

Regarding the inter-cluster distance, the maximum genetic distance was observed between the clusters II and V (23.44) followed by clusters I and V (17.56), while the minimum was observed between the clusters III and IV (7.29) followed by cluster I and II (7.48). The maximum value of inter-cluster distance indicated that the genotypes belonging to cluster V was far diverged from those of cluster II. The minimum inter-cluster divergence was observed between cluster IV and III (7.29) indicating that the genotypes of these clusters were genetically closed. However, genotypes within the other pair of clusters indicated that they were less diverged. The inter-cluster distances in all the clusters were higher than the intra-cluster distances suggesting wider genetic diversity among the genotype of different groups. The results were in agreement with Islam *et al.* (2016), Akter *et al.* (2016), Ahmed *et al.* (2015b), Islam *et al*. (2014) and Sohrabi *et al.* (2012).

Table 7 presents the cluster-mean values for all the 15 quantitative characters. The data revealed that different clusters exhibited the highest and lowest mean values of individual characters and none of the single cluster showed the highest or lowest mean values of all the characters. However, the highest cluster means for yield/hill, days to maturity and effective tiller number were obtained from cluster III. The highest flag leaf length, leaf area index, filled grains per panicle were found in cluster V while the lowest culm diameter, short duration, low yielding genotypes were clubbed into cluster II. Dwarf stature and lower flag leaf length were recorded into cluster I whereas the highest plant height and panicle length were obtained from cluster IV. Maximum good characters were accumulated in cluster III and as a result

Table 6. Intra (bold) and inter-cluster distances (D^2) for 30 Jhum rice landraces.

Cluster	I	II	III	IV	V
I	**0.57**	7.48	12.20	8.94	17.56
II		**0.85**	17.10	11.54	23.44
III			**1.62**	7.29	7.51
IV				**0.68**	13.24
V					**0.77**

Table 7. Cluster means for 15 quantitative characters in 30 Jhum rice landraces.

Character	I	II	III	IV	V
Flag leaf length (cm)	38.47	38.96	40.27	45.20	46.17
Flag leaf width (cm)	1.75	1.57	1.53	1.56	1.61
Leaf area index (cm^2)	50.42	46.20	46.39	52.65	55.56
Culm diameter (mm)	4.69	4.42	5.05	4.47	5.22
Plant height (cm)	109.43	113.68	118.20	131.31	122.87
Effective tiller number	7.00	7.00	9.00	8.00	6.00
Panicle length (cm)	26.83	28.16	28.63	29.60	28.73
Days to flowering	91	86	94	89	91
Days to maturity	119	114	120	116	118
Filled grains per panicle	89.00	62.00	134.00	106.00	160.00
Grain length (mm)	9.64	9.69	8.22	7.79	8.48
Grain breadth (mm)	3.16	3.24	2.87	3.05	3.15
Length-breath ratio	3.10	3.04	2.96	2.58	2.78
TGW (g)	28.42	28.00	19.15	22.13	23.13
Yield/hill (g)	5.12	4.41	6.82	5.26	4.83

higher yield (6.82 g/hill) was also obtained in this cluster. But it was interesting that in most of the cases cluster V could produced the highest D^2 values with all other clusters except cluster III. So they can be used in hybridization programme to produce higher yielding genotypes. Therefore, the landraces under cluster II, cluster III and cluster V might be selected for future breeding programme. Siddique *et al*. (2013, 2016) and Sohrabi *et al*. (2012) earlier reported similar trend of conclusions on rice using Mahalanobis' D^2 statistics.

Table 8 presents contributions of the characters towards divergence. The canonical variate analysis revealed that the vectors (Vector I and II) for culm diameter, days to flowering, days to maturity and length-breath ratio were positive. Such results indicated that these four characters contributed maximum towards divergence. It is interesting that the greater divergence in the present materials due to these four characters will offer a good scope for improvement of yield through rational selection of parents for producing heterotic rice hybrids. Similar findings also reported by Islam *et al.* (2009) for local rice germplasm.

Table 8. Relative contributions of the fifteen characters to the total divergence in Jhum rice landraces.

Character	Vector 1	Vector 2
Flag leaf length (cm)	0.300	-0.068
Flag leaf width (cm)	0.125	-0.017
Leaf area index (cm^2)	0.311	-0.057
Culm diameter (mm)	0.294	0.177
Plant height (cm)	0.272	-0.250
Effective tiller number	-0.103	-0.219
Panicle length (cm)	0.292	-0.190
Days to flowering	0.407	0.215
Days to maturity	0.396	0.218
Filled grains per panicle	0.164	-0.225
Grain length (mm)	-0.068	0.513
Grain breadth (mm)	-0.275	-0.151
Length-breath ratio	0.133	0.450
TGW (g)	-0.293	0.318
Yield/hill (g)	0.0735	-0.280

CONCLUSIONS

Based on the results of the current study, we made some conclusions as follows: (i) Jhum rice landraces from Rangamati district showed good diversity in both qualitative and quantitative agro-morphological traits. (ii) Cluster analysis using quantitative traits of

30 landraces grouped them into five clusters. (iii) Observed variances in qualitative data were mostly explained by blade pubescence, anthocyanin colour of leaf sheath, apiculus colour, stigma colour, lemma-palea colour, grain tip colour and awning while considering results from genetic distance, the landraces Koborok -2, Chorui, Galong, Bidi, Kamarang-1 and Suri could be selected as parents for further breeding programme. Besides, selection of parents from the clusters II and V may be useful for breeding to obtain desirable transgressive segregants. Moreover, characterizations of the identified landraces need to be done using SSR markers for protection from biopiracy.

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DNA Fingerprinting and Genetic Diversity in Aus Rice (*Oryza sativa* L.) Landraces of Bangladesh

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ABSTRACT

The allelic diversity and relationships among 48 Aus rice landraces were determined through DNA fingerprinting using microsatellite (SSR) markers. A total of 14 SSR markers for different chromosomes were used to characterize and differentiate the studied rice genotypes. The number of alleles per locus varied from three alleles (RM118) to 18 alleles (RM44) with an average of 9.88. The polymorphic information content (PIC) varied widely among the loci and ranged from 0.3725 (RM107) to 0.9146 (RM519) with an average of 0.7248. The genetic distance-based results found in the UPGMA clustering system revealed six genetic groups with a similarity coefficient of 0.35. Chakila and Shitki saitta had closest distance in the SSR based genetic distance might have same genetic background. Based on genetic coefficient, the diverse landraces Kasalot, Balam, Pankhiraj, Dular, Hashikalmi, Galong, Panbira, Marichbati, Pidi and Surjomoni could be selected as potential parents for varietal improvement programme. The findings of this study should be useful for varietal identification and could be useful for plant breeders in selecting suitable genetically diverse parents for the crossing programme.

Key words: Aus rice, DNA fingerprinting, genetic diversity, SSR markers, UPGMA clustering

INTRODUCTION

Rice (*Oryza sativa* L.) is one of the most important food crops and a primary source of food for more than half of the world's population (Khush, 2005). It has been cultivated in Asia since ancient times and for generations farmers have maintained thousands of different landraces (Jackson, 1995). Bangladesh is already under pressure both from huge and increasing demands for food, and from problems of agricultural land and water resources depletion. Bangladesh needs to increase the rice yield in order to meet the growing demand for food emanating from population growth. More than 1,27,000 rice accessions and wild relatives can be found in the world's largest Genebank for rice at IRRI (International Rice Research Institute) based in the Philippines (http://irri.org/our-work/research/genetic-diversity). Until now, Bangladesh Rice Research Institute (BRRI) has collected and preserved more than 8,000 varieties/landraces/cultivars/wild types from indigenous and exotic sources in the genebank. Out of them, 8,200 have been registered.

For the assessment of plant genetic diversity, the number of scoreable morphological characters is varying as compared to the biological active genes. Moreover, in most cases, plant genomes have large amount of repetitive DNA, which are not expressed and do not contribute to the physiological or morphological appearance of plants. In case of very closely related plant varieties, there are very few morphological differences, which as a matter of fact do not represent the true genetic differences at DNA level. So, there is always a need to study polymorphism at DNA level, which could be an indicator of genetic diversity. Several types of molecular markers *viz* RFLP, RAPD, AFLP, microsatellites (SSR) and SNP have been developed. PCR-based markers such as simple sequence repeat (SSR) are co-dominant, hyper variable, abundant and well distributed throughout the rice genome (Temnykh *et al*., 2001). Microsatellites have shown great promise in genetic diversity, genome mapping, gene tagging and marker-assisted selection (MAS) because they are technically simple, time saving, highly

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informative and require small amount of DNA. Microsatellite markers are now available through the published high-density linkage map (McCouch *et al*., 2002; IRGSP, 2005) or public database. A study was conducted on 234 rice landraces in Plant breeding division, Cornell University and they identified five distinct groups corresponding to indica, aus, aromatic, temperate japonica and tropical japonica rice (Amanda *et al*., 2004). They also have very high diversity with 98% of loci polymorphic Aus group. Despite their drought tolerance and early maturity, the group has received less attention compared to indica and japonica group.

There are four distinct ecotypes of rice- Boro, Aus, Transplanted Aman and Deep water Aman in Bangladesh. Bangladesh has a good source of indigenous rice cultivars comprising Boro, Aman and Aus landraces. Those landraces have good adaptation having poor yield. Actually cultivation of these landraces was gradually replaced by high yielding varieties during the last 20 years. These landraces adapted in different parts of the country, some of which have very nice quality, fineness, aroma, taste and high protein contents (Dutta *et al*., 1998). After establishment of BRRI, DNA fingerprinting has been done only of a small number of local landraces. Indigenous rice landraces were characterized at both molecular and phenotypic level by many countries. They have been done for their crop identity and searched for new genes for further crop improvement. However, information on the genetic diversity of local landraces particularly for Aus rice is very scanty in Bangladesh. Precise information on the extent of genetic diversity among population is crucial in any crop improvement programme, as selection of plants based on genetic diversity has become successful in several crops (Ananda and Rawat, 1984; De *et al*., 1988). Hence, the objectives of this research were to (1) assess the genetic variation and diversity of 48 Aus rice genotypes, (2) determine the genetic relationship among these genotypes for breeding purposes, and (3) characterize these rice genotypes at molecular level (DNA fingerprinting) that would help in the establishment of varietal rights.

MATERIALS AND METHODS

Plant materials

Forty-eight Aus rice landraces were used in this study (Table 1). A five gram seed from each of the entry was germinated and then sown in earthen pots for growth and subsequent DNA extraction.

SSR markers

Fourteen SSR markers were selected (Table 2) to detect DNA for discriminating the tested Aus rice landraces.

Genotyping protocol

Total genomic DNA was extracted from young leaves of three-week-old plants following the simple and modified protocol of Zheng *et al*., 1995. PCR analysis was performed in 12.5 µl reaction sample containing 5-25 ng of DNA template, 1.25 µl of $MgCl_2$ free 10X PCR buffer (100 mM Tris-HCl pH 9.0 at 25°C, 500 mM KCl, 0.1% Triton® X-100 and H_2O), 1.5 µl of 25 mM $MgCl_2$, 0.25 µl of 10mM dNTP, 0.25 µl of 5 U/µl Taq polymerase enzyme, 0.625 µl each of 10 µM forward and reverse primers using a MJ Research single 96-well thermal cycler. The mixture was overlaid with one drop of mineral oil to prevent evaporation. After initial denaturation for five minutes at 94°C, each cycle comprised one min denaturation at 94°C, one min annealing at 55°C, and two min extension at 72°C with a final extension for sevem min at 72°C at the end of 35 cycles. The PCR products were mixed with bromophenol blue gel loading dye and were analyzed by electrophoresis on 8% polyacrylamide gel using mini vertical polyacrylamide gels for high throughput manual genotyping (CBS Scientific Co. Inc., CA, USA). 2.5 µl of amplification products were resolved by running gel in 1×TBE buffer for 2-2.5 hrs depending upon the allele size

Table 1. Rice landraces used in the present study with their provenance.

Landrace	BRRI accession no.	Place of collection	Landrace	BRRI accession no.	Place of collection
Kasalot	3283	Sylhet	Panbira	50	Dhaka
Balam	4945	Khulna	Hasha kumira	63	Manikganj
Kamranga	4926	Rangamati	Dhala saita	64	Manikganj
Surjomoni	3550	Barisal	Loroi	65	Dhaka
Gopalbogh	2110	Narsingdi	Boteswar	66	Dhaka
Haitta	3232	Tangail	Chakila	152	Tangail
Goarchara	953	Khulna	Shitki saita	154	Tangail
Chengri	808	Sylhet	Baila bokri	155	Tangail
Saita	1052	Jessore	Kataktara (2)	183	Mymensingh
Company	7270	Khagrachhari	Bolorum	188	Mymensingh
Galon	743	Chittagong	Pankiraj	189	Mymensingh
Kamarang	7263	Khagrachhari	Hanpa	190	Mymensingh
Maloti	7269	Khagrachhari	Agaua	191	Mymensingh
Lal binni	753	Chittagong	Chakulia	193	Mymensingh
Suri dhan	7264	Khagrachhari	Shala dumra	263	Rangpur
Galong	7265	Khagrachhari	Sukhti	264	Rangpur
Sada galon	4923	Rangamati	Faisha manja	265	Rangpur
Sili	7251	Bandarban	Inda	266	Rangpur
Mongthong	7249	Bandarban	Thubri	267	Rangpur
Kilong	7257	Bandarban	Bolun	268	Rangpur
Dharial	18	Dhaka	Phul dumra	269	Rangpur
Dular	22	Dhaka	Kola dama	271	Rangpur
Hashikalmi	30	Dhaka	Turki	7423	Khagrachhari
Marichbati	47	Dhaka	Pidi	7770	Bandarban

Table 2. List of the 14 SSR markers.

Locus name	Chr. no.	Repeat motif	Forward primer	Reverse primer
RM6	2	(AG)16	GTCCCCTCCACCCAATTC	TCGTCTACTGTTGGCTGCAC
RM204	6	(CT)44	GTGACTGACTTGGTCATAGGG	GCTAGCCATGCTCTCGTACC
RM241	4	(CT)31	GAGCCAAATAAGATCGCTGA	TGCAAGCAGCAGATTTAGTG
RM279	2	(GA)16	CCTCTCACTCACGTGGACTCTCC	CCTCACCCTAGGCTTTGATATGC
RM519	12	(AAG)8	AGAGAGCCCCTAAATTTCCG	AGGTACGCTCACCTGTGGAC
RM286	11	(GA)16	GGCTTCATCTTTGGCGAC	CCGGATTCACGAGATAAACTC
RM44	8	(GA)16	ACGGGCAATCCGAACAACC	TCGGGAAAACCTACCCTACC
RM147	10	(TTCC)5(GGT)5	TACGGCTTCGGCGGCTGATTCC	CCCCCGAATCCCATCGAAACCC
RM107	9	(GA)7	AGATCGAAGCATCGCGCCCGAG	ACTGCGTCCTCTGGGTTCCCGG
RM118	7	(GA)8	CCAATCGGAGCCACCGGAGAGC	CACATCCTCCAGCGACGCCGAG
RM10	7	(GA)15	TTGTCAAGAGGAGGCATCG	CAGAATGGGAAATGGGTCC
RM161	5	(AG)20	TGCAGATGAGAAGCGGCGCCTC	TGTGTCATCAGACGGCGCTCCG
RM1	1	(GA)26	GCGAAAACACAATGCAAAAA	GCGTTGGTTGGACCTGAC
RM85	3	(TGG)5(TCT)12	CCAAAGATGAAACCTGGATTG	GCACAAGGTGAGCAGTCC

Chr.=Chromosome.

at around 75 volts and 180 mA current. The gels were stained in 0.5 mg/ml ethidium bromide and were documented using UVPRO (Uvipro Platinum, EU) gel documentation unit. Microsatellite or simple sequence repeat (SSR) markers were used for DNA analysis (Temnykh *et al*., 2001; McCouch *et al*., 2002).

Data analysis

Size for each amplified allele was measured in base pair using Alpha-EaseFC 5.0 software. The summary statistics including the number of alleles per locus, major allele frequency, gene diversity, polymorphism information content (PIC) values were determined using PowerMarker version3.25 (Liu and Muse, 2005). The allele frequency data from Power Marker was used to export in binary format (allele presence=1 and allele absence=0) for analysis with NTSYS-pc version 2.1 (Rohlf, 2002). A similarity matrix was calculated with the Simqual subprogramme using the Dice coefficient, followed by cluster analysis with the SAHN subprogramme using the UPGMA (unweighted pair group method using arithmetic mean) clustering method as implemented in NTSYS-pc.

RESULTS AND DISCUSSION

Overall SSR diversity

Forty-eight Aus landraces were successfully amplified with the 14 SSR marker pairs where marker pairs referred to as loci and DNA bands as alleles. A total of 138 alleles were detected using 14 microsatellite markers across 48 rice landraces. The number of alleles per locus ranged from three alleles (RM118) to 18 alleles (RM44), with an average of 9.88 (Table 3). PIC values ranged from 0.3725 (RM107) to 0.9146 (RM519) with an average of 0.7248. The PIC values for other markers were 0.9135 (RM286), 0.9115 (RM44), 0.8620 (RM147), 0.8439 (RM6), 0.8247 (RM279), 0.7961 (RM241), 0.7371 (RM85), 0.7252 (RM204), 0.6867 (RM1), 0.6251 (RM10), 0.5301 (RM161) and 0.4038 (RM118) respectively. The allele frequency ranged from 12.50% (RM519, RM286, RM44) to 77.08% (RM107) with an average of 35.12 (Table 3). PIC value revealed RM519 as the best marker. Figugre 1 shows the DNA profiles of 48 Aus rice landraces with SSR marker RM519.

Genetic distance-based analysis

The SSR marker data were subjected to cluster analysis using NTSYS programme. The similarity matrix was constructed using Dice coefficient method. Cluster analysis was done to group the genotypes into a dendrogram. From this dendrogram, the 48 rice landraces were grouped into six major clusters at a coefficient of 0.35 and the similarity coefficient value ranged from 0.10 to 0.86. Cluster I consisted of 18 accessions and is the biggest group among six clusters, followed by cluster IV which contained 11 accessions; cluster V comprised nine accessions; cluster II comprised six accessions; cluster III and VI were both composed of two accessions (Fig. 2).

In rice improvement breeding programme the genetically diverse landraces could be chosen as parents for crossing programme to create genetic variability and can be used as transgressive segregants. It was found that two Aus landraces (Chakila and Shitki saita) were sorted out as exactly same genotypes in this analysis might possess same genetic background. Hence, microsatellite marker based molecular fingerprinting could serve as a potential basis in the identification of genetically distant accessions as well as in duplicate sorting of the morphologically close accessions.

In the present investigation, the SSR markers generated 138 alleles with the number of alleles per locus varying from 3 to 18. Similar number of microsatellite markers previously used as subset for genetic diversity analysis of *O. sativa* (Siddique *et al*., 2016a). The average number of alleles per locus was 9.88, indicating a greater magnitude of diversity among the rice landraces. This value is comparable to five alleles (RM275) to 15 alleles (RM180), with an average of 9.7 across the 30 loci (Siddique *et al.*, 2016b). The PIC values

Table 3. Allele number, allele size, allele frequency and PIC of 48 Aus rice landraces for 14 SSR markers.

Marker	Chr. no.	Position (cM)	Allele no.	Allele size range (bp)	Allele freq (%)	PIC value
RM6	2	29.57	12	142-178	27.08	0.8439
RM204	6	24.04	8	106-118	39.58	0.7252
RM241	4	26.82	12	101-127	33.33	0.7961
RM279	2	13.2	11	119-136	27.08	0.8247
RM519	12	19.90	16	117-145	12.50	0.9146
RM286	11	0.38	15	95-123	12.50	0.9135
RM44	8	2.88	18	97-122	12.50	0.9115
RM147	10	20.68	14	87-109	27.08	0.8620
RM107	9	20.06	6	165-187	77.08	0.3725
RM118	7	28.01	3	163-166	52.08	0.4038
RM10	7	6.50	4	156-148	43.75	0.6251
RM161	5	27.89	4	163-189	43.75	0.5301
RM1	1	4.63	7	80-90	41.67	0.6867
RM85	3	66.76	8	89-117	4167	0.7371
Mean			10		35.12	0.7248

Motif of the SSR and number of repeats as previously published (http://www.gramene.org).

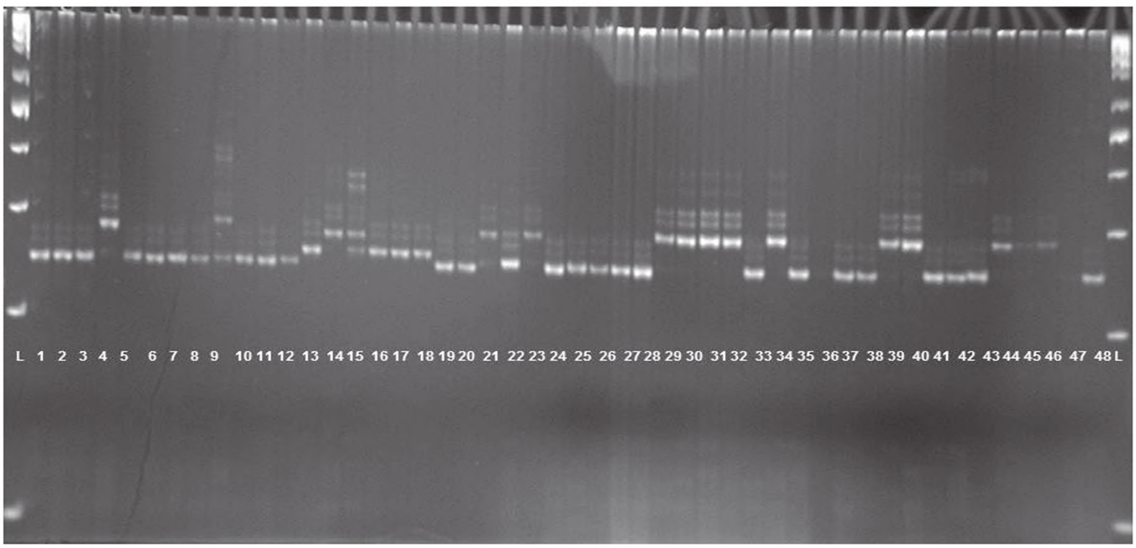

Fig. 1. DNA profile of 48 Aus rice landraces with RM519.

Legend: Lane 1=Kasalat, 2=Balam, 3=Kamranga, 4=Surjomoni, 5=Gopalbogh, 6=Haitta, 7=Goarchara, 8=Chengri, 9=Saita, 10=Company, 11=Kamarang, 12=Maloti, 13=Lalbinni, 14=Suridhan, 15=Galong, 16=Sada galon, 17=Sili, 18=Mongthong, 19=Kilong, 20=Dharial, 21=Dular, 22=Hashikalmi, 23=Marichbati, 24=Panbira, 25=Hasha kumira, 26=Dhala saita, 27=Loroi, 28=Boteswar, 29=Chakila, 30=Shitki saita, 31=Baila bokri, 32=Kataktara (2), 33=Bolorum, 34=Pankiraj, 35=Hanpa, 36=Agaua, 37=Chakulia, 38=Shala dumra, 39=Sukhti, 40=Faisha manja, 41=Inda, 42=Thubri, 43=Bohum, 44=Phul durma, 45=Kola dama, 46=Bangal bakri, 47=Turki, 48=Pidi.

ranged from 0.37 (RM107) to 0.91 (RM519, RM286, RM44) with an average of 0.7248. The PIC values observed, are comparable to previous estimates of microsattelite analysis in rice *viz.* 0.34-0.88 (Thomson *et al.*, 2007), 0.65-0.91 (Siddique *et al.*, 2014), 0.59-0.90 (Siddique *et al.*, 2016b). The alleles revealed by markers showed a high degree of polymorphism. The

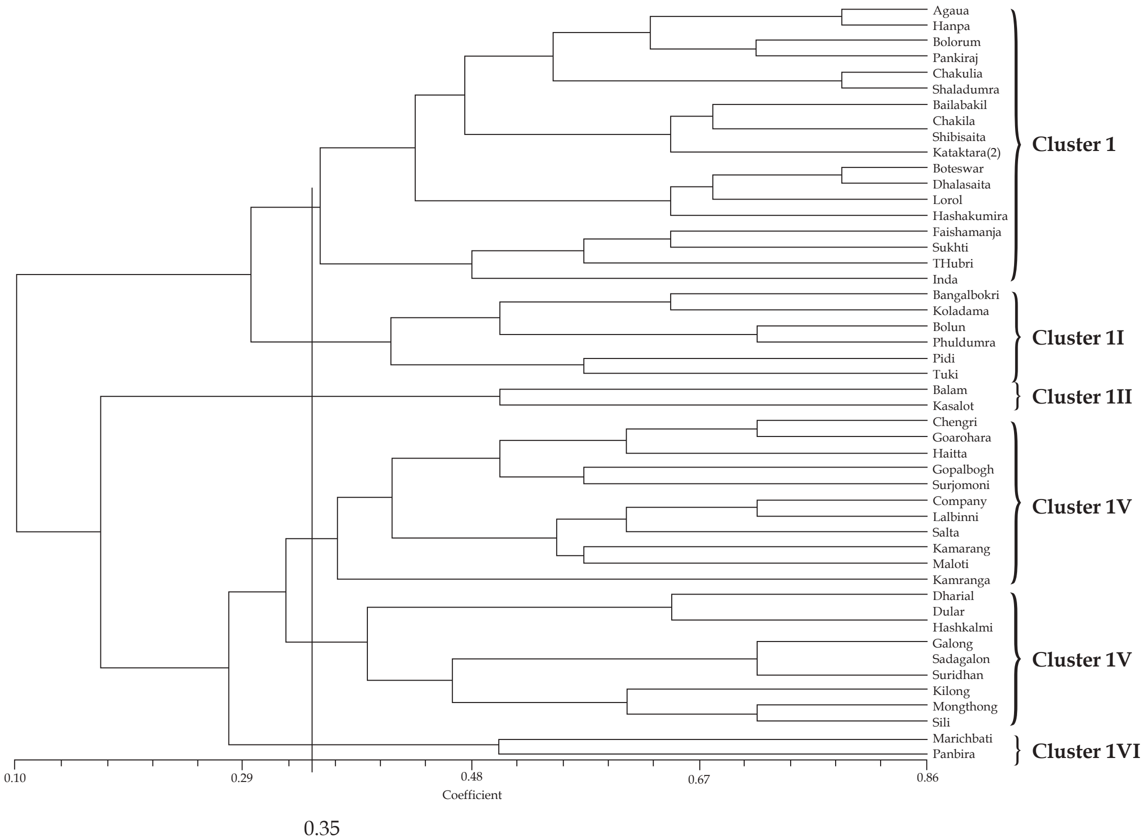

Fig. 2. An UPGMA eluster dendogram showing the genetic relationship among 48 Aus landrances based on the alleles detected by 14 SSR markers.

mean PIC value observed in this study was higher than the PIC value of 0.578 recorded by Ravi *et al*., (2003) in an earlier study among rice cultivars, landraces and wild relatives. This could indicate that the genotypes used in the present study were more diverse. Panaud *et al*., (1996) described high genetic similarity among landraces of common geographic origin and low similarity among landraces of diverse geographic origins using SSR markers in rice.

CONCLUSIONS

The results obtained from this study provided some useful implications for establishment of Intellectual Property Rights (IPR) issues of Bangladeshi rice landraces. There was a high level of genetic diversity among accessions of Aus rice landraces, suggesting that SSR markers were effective in the detection of polymorphism in this ecosystem. To broaden the genetic base and for the improvement of Aus rice, landraces having the lowest genetic similarities could be selected as parents. Therefore, hybridization should be made between two distant populations. Considering all these criteria and results, the diverse landraces Kasalot, Balam, Pankhiraj, Dular, Hashikalmi, Galong, Panbira, Marichbati, Pidi and Surjomoni could be selected as parents for further breeding programmes. This will bring about greater diversity, which will lead to a high productive index in terms of increase in yield and overall quality.

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Sensitivity of Annual Weeds against Sulfentrazone 48 SC herbicide in Rice Cultivation

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ABSTRACT

Herbicides are used successfully for weed control in rice fields for rapid effect, easier to application and low cost involvement in comparison to the traditional methods of hand weeding. Sulfentrazoneis a new pre-planting herbicide in Bangladesh. Field trials were conducted at Bangladesh Rice Research Institute (BRRI), Gazipur during Aman 2014 and Boro 2014-15 to evaluate the efficacy of Sulfentrazone 48 SC on weed suppression and performance of transplanted rice. Treatments were Sulfentrazone 48 SC @ 150, 200 and 250 ml ha^{-1}, pretilachlor 500 EC @ 1000 ml ha^{-1}, weed free and unweeded control. Visual assessment indicated that this herbicide possesses high selectivity and not toxic to rice plants. The results revealed that the major weed flora associated with the transplanted rice was mainly comprised of two grasses, two sedges and four broadleaves in Aman 2014 and two grasses, two sedge and two broadleaves in Boro 2014-15. The most dominant weeds were *Cyperus difformis, Echinochloa crus-galli, Scirpus maritimus* and *Monochoria vaginalis* in both the growing seasons. Application of Sulfentrazone 48 SC @ 200 ml ha^{-1} was most effective to suppress weed density and dry masses in both the seasons resulting increased grain yield more than 50% as compared to unweeded control. Therefore, Sulfentrazone 48 SC @ 200 ml ha^{-1} should apply two or three days before plantingfor effectively control weeds in rice.

Key words: Sulfentrazone, transplanted rice, weed density, weed control efficiency

INTRODUCTION

Among the cereals, rice (*Oryza sativa* L.) is the most important and extensively grown in tropical and subtropical regions of the world, and staple food for more than 60 per cent of the world population. The average yield of rice in Bangladesh is 4.5 t ha^{-1} (BRRI, 2016). Rice production needs to be increased by 50% or more above the current production level to meet the rising food demand (Sunyob *et al*., 2015). Weed infestation and interference is a serious problem in rice fields that significantly decreases yield of rice. In Bangladesh weed infestation reduces rice grain yield by 70-80% in Aus, 30-40% in transplanted Aman and 22-36% in modern Boro rice cultivars (BRRI, 2006; Mamun, 1990). Losses due to infestation of weeds are greater than the combined losses caused by insect, pest and diseases in rice (Willocquet *et al*., 1998 and Bari, 2010). Weeds not only cause huge reductions in rice yields but also increase cost of cultivation, reduce input efficiency, interfere with agricultural operations, impair quality, act as alternate hosts for several insect pests, diseases. They affect aesthetic look of the ecosystem as well as native biodiversity, human and cattle health. Weed competes for nutrient, space, sunlight and consume the available moisture with crop plant resulting in crop yield reduction (Sunyob *et al*., 2015). Weed infestation in rice cultivation is a major constraint and expensive as well. Since hand weeding including all other weed controlling methods still are not easy so that uses of chemicals are the obvious and cost efficient (Jayadeva, 2010). Now-a-days chemical control of weed has become popular in Bangladesh due to scarcity of labour during peak growing season, and lower cost involvement. In Bangladesh, the annual consumption of herbicides grew over 3,420 metric tons in 2014 (BCPA, 2016) compared to only 108 metric tons during 1986-87 (BBS, 1991), and the growth is almost

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exponential. The traditional methods of weed control practices in Bangladesh are preparatory land tillage, hand weeding by hoe and hand pulling. Mechanical and cultural weed control in transplanted rice is an expensive method. Especially at the peak period of labour crisis sometimes weeding becomes late resulting drastic yield loss. Abundant use of pre and post emergence herbicides such as butachlor, pretilachlor, oxadiazone, pyrazosulfuron ethyl, ethoxysulfuron alone or supplemented with one hand weeding found effective for weed management in transplanted rice (Bhuiyan, 2016). Sulfentrazone has been recently released for pre planting weed control in rice field in Bangladesh. It belongs to Triazolinones chemical group. Mode of action is the destruction of cell membranes by inhibiting the enzyme Protox and consequently the destruction of cell membranes. The chemical formula of sulfentrazone is 4′-dichloro-5′-(4-difluoromethyl-4,5-dihydro-3-methyl-5-oxo-1H-1,2,4-triazol-1-yl) methane sulfonanilide; N-[2,4-dichloro-5-[4-(difluoromethyl)-4,5-dihydro-3-methyl-5-oxo-1H-1,2,4-triazol-1-yl] phenyl] methane sulfonamide. It is a selective herbicide, absorbed mainly by the shoots of germinating plants. It can effectively control most important perennial and annual species of Broadleaf weeds, grasses and sedges in transplanted rice. Furthermore this type of herbicide is almost new adding up in Bangladesh for control of weeds before planting. Therefore, to provide a wider choice of effective herbicide for farmers there is a crucial need to develop environmental friendly molecules of newer chemistries with different mode of action. Considering the situation, the present study was undertaken to evaluate the efficacy of sulfentrazone for annual weed suppression and to find out an appropriate dose and its impacts on transplanted rice.

MATERIALS AND METHODS

The experiment wasconducted at Bangladesh Rice Research Institute, Gazipur, situated at 23°59´33´´ N and 90°24´19´´ E at an elevation of 8.4 m from the mean sea level, and is characterized by sub-tropical climate during Aman 2014 and Boro 2014-2015 seasons to evaluate the efficacy of sulfentrazone for weed suppression and to find out an appropriate dose of this herbicide and its impacts on transplanted rice. The commercial name of sulfentrazone is Authority 48 SC. The soil of the experimental field was non-calcareous dark grey flood plain (FAO, 2004) with pH around 6.2 and low in organic matter (1.2%). The experiment was carried out with six treatments. Table 1 presents details of experimental treatments and table 2 presents details of herbicides. All treatments were laid out in a randomized complete block design with three replications. Twenty-five-day-old seedlings of BRRI dhan49 for T. Aman 2014 and 35-day-old seedlings of BRRI dhan28 for Boro 2014-2015 were transplanted. Two/three seedlings per hill were transplanted maintaining 20 × 20 cm spacing. The field was fertilized following BRRI recommendation (T. Aman: N:P:K:S= 69:10:41:11 kg ha^{-1} and Boro; N:P:K:S= 120:19:60:24 kg ha^{-1}) (BRRI, 2013). Herbicides were sprayed three days before transplanting with the help of a knapsack sprayer. In weed free treatment, the plots were kept weed free up to 45 DAT by hand weeding and check herbicide was Pretilachlor (commercial name is Rifit 500 EC). Authority 48 SC (Sulfentrazone) herbicide is new in Bangladesh and its phytotoxicity needs to be evaluated on rice crop. The phytotoxicity of the herbicide onrice plants was determined by visual observations (yellowing leaves, burring leaf tips, stunting growth etc). The degree of toxicity on rice plant was measured by the following scale used by IRRI (1965):

Rating	Symptom
1	No toxicity
2	Slightly toxicity
3	Moderate toxicity
4	Severe toxicity
5	Toxic (plant killed)

Table 1. Weed management treatments of the experimental plots.

Label	Treatment	Rate of application	Application time of herbicide / Operation of hand weeding
T_1	Authority 48 SC (Sulfentrazone) + 1 HW on 45 DAT	150 ml ha^{-1}	3 days before transplanting
T_2	Authority 48 SC (Sulfentrazone) + 1 HW on 45 DAT	200 ml ha^{-1}	3 days before transplanting
T_3	Authority 48 SC (Sulfentrazone) + 1 HW on 45 DAT	250 ml ha^{-1}	3 days before transplanting
T_4	Rifit 500 EC (Pretilachlor)	1000 ml ha^{-1}	3 days after transplanting
T_5	Weed free by three hand weeding	-	15, 30 and 45 days after transplanting
T_6	Unweeded (Control)	-	No weeding

DAT= Days after transplanting.

Table 2. Details of herbicide.

Trade name	Active ingredients (a. i.) rate	Name of active ingredient	Chemical name	Chemical family
Authority 48 SC	75 ml ai ha^{-1}	Sulfentrazone	2′, 4′-dichloro-5′-(4-difluoromethy l-4, 5-dihydro-3-methyl-5-oxo-1H-1, 2, 4-triazol-1-yl) methane sulfonanilide; N-[2, 4-dichloro-5-[4-(difluoromethyl)-4, 5-dihydro-3-methy l-5-oxo-1H-1, 2, 4-triazol-1-yl]phenyl] methane sulfonamide	Triazolinones
Authority 48 SC	96 ml ai ha^{-1}	Sulfentrazone	do	do
Authority 48 SC	120 ml ai ha^{-1}	Sulfentrazone	do	do
Rifit 500 EC	500 ml ai ha^{-1}	Pretilachlor	2-chloro-2′,6′-diethyl-N-(2-propoxyethyl) acetanilide	Chloroacetamide

The rating of toxicity was done within 15 days after application of herbicides. It was observed four times- at 6, 9, 12 and 15 days after application and the mean rate was calculated from 10 sample plants of a unit plot.

Data on weed density and dry weight were taken from each plot on 40 DAT. The weeds were identified species-wise. Dry weights of weeds were taken by drying them in electric oven at 60°C for 72 hours followed by weighing by digital balance. Relative weed density (RWD), relative weed biomass (RWB) and weed control efficiency (WCE) of different weed control treatments were calculated with the following formulas (Tabib *et al*., 2013 and 2014):

$$\text{RWD (\%)} = \frac{\text{Density of individual weed species in the community}}{\text{Total density of all weed species in the community}} \times 100$$

$$\text{RWD (\%)} = \frac{\text{RWD (\%)} + \text{RWB (\%)}}{2} \times 100$$

$$\text{WCE (\%)} = \frac{\text{Dry weight of weed in weedy check plots-Dry weight of weeds in treated plots}}{\text{Dry weight of weeds in weedy check plots}}$$

Data on panicle m^{-2}, grains panicle^{-1}, 1000 grain weight (TGW), sterility (%) and grain yield were collected according to the standard protocol. Analysis of Variance of the measured parameters were analyzed and graphical presentation were done by using STAR 2.0.1 software.

RESULTS AND DISCUSSION

Phytotoxicity of herbicides on rice plant

Table 3 presents the degree of toxicity of the herbicide to rice plants and the symptoms produced on plant are. It is observed that Authority 48 SC @ 150 ml ha^{-1} showed no toxicity and Authority 48 SC @ 200 ml ha^{-1} showed very slight yellowing of leaves while Authority 48 SC @ 250 ml ha^{-1} showed moderate yellowing of leaves. Phytotoxicity symptoms observed not more prominent for using this herbicide. Phytotoxicity of rice plant by combined herbicide resulted less which is similar to the findings of Bhuiyan *et al*., 2010.

Weed infestation during T. Aman season 2014

The rice field was infested with different types of weeds. The relative density of these weed species was also dissimilar (Table 4). Eight different weeds species were observed in unweeded (control) plot where most dominating weeds were sedges and broadleaves. Among the infestation of different categories of weeds, two were grasses, two sedges and four broadleaves. The weed species were belonged to the families of Poaceae, Cyperaceae, Pontederiaceae, Marsileaceae, Sphenocleaceae and Asteraceae. The broadleaf weeds were: *Monochoria vaginalis*, *Marsilea minuta*, *Sphenoclea zeylanica* and *Eclipta alba*; grasses were: *Echinochloa crus-galli*, *Cynodon dactylon*; and sedges were *Cyperus difformis* and *Scirpus maritimus*. Among the weed species maximum relative weed density (RWD) was observed for *Cyperus difformis* (31.24%) followed by *Echinochloa crus-galli* (30.40%). However, highest relative weed biomass (RWB) was observed for *Echinochloa*

Table 3. Rating of herbicide toxicity on rice plant under different treatments.

Treatment	Rating		Symptom
	Boro 2014-15	T. Aman 2015	
Authority 48 SC @ 150 ml ha^{-1} (72 ml a.i. ha^{-1})	1.10	1.10	No toxicity
Authority 48 SC @ 200 ml ha^{-1} (96 ml a.i. ha^{-1})	1.20	1.17	Sometimes slight yellowing of leaves
Authority 48 SC @ 250 ml ha^{-1} (120 ml a.i. ha^{-1})	2.10	2.10	Slight yellowing of leaves which required 5-7 days to recover
Rifit 500 EC @ 1000 ml ha^{-1} (500 ml a.i. ha^{-1})	1.10	1.15	No toxicity

Table 4. Weed composition, relative weed density (RWD) and relative weed biomass (RWB) in the untreated control plots in Aman 2014 at BRRI, Gazipur.

Weed species	Family	Class	RWD (%)	RWB (%)
Cynodon dactylon	Poaceae	Grass	7.65	9.63
Echinochloa crus-galli	Poaceae	Grass	30.40	32.75
Cyperus difformis	Cyperaceae	Sedge	31.24	32.45
Scirpus maritimus	Cyperaceae	Sedge	24.76	23.92
Monochoria vaginalis	Pontederiaceae	Broadleaf	22.28	27.29
Marsilea minuta	Marsileaceae	Broadleaf	11.47	12.64
Sphenoclea zeylanica	Sphenocleaceae	Broadleaf	3.81	2.58
Eclipta alba	Asteraceae	Broadleaf	2.84	3.45

crus-galli (32.75%) followed by *Cyperus difformis* (32.45%). Among the weeds, *Eclipta alba* was minor weed with 2.84% RWD and 3.45% RWB. It was observed that broadleaf weeds were less dominating species. Bhuiyan (2016) explained that efficacy and weed infestation of rice plant depends on different types of herbicides used.

Weed infestation during Boro season 2014-15

The number of infesting weed species was slightly different in Boro season than in T. Aman season. These weed flora were ecologically categorized into two broadleaves, two sedges and two grasses (Table 5). The major weed was *Cyperus difformis* of which relative weed density (RWD) and relative weed biomass (RWB) was 32.50% and 34.07%, respectively. The second dominant weed was *Echinochloa crus-galli* of which RWD was 31.48% and relative weed biomass (RWB) was 33.76%. In Boro season broadleaf weeds created less dominance than in T. Aman season. Authority 48 SC effectively control *Echinochola* and *Cyperus* sp. which was similar to the findings of Puniya *et al*., 2007.

Weed ranking

The summed dominance ratio (SDR) is an important indicatorto showthe ranking of weeds in a community. The most dominant weeds in T. Aman 2014 were *Cyperus difformis, Echinochloa crus-galli, Scirpus maritimus* and *Monochoria vaginalis* (Fig. 1). *Cyperus difformis, Echinochloa crus-galli, Scirpus maritimus* and *Monochoria vaginalis* were also the most dominant weeds in Boro 2014-15. Mamun *et al*., 2011 showed that SDR of a weed against same herbicide was almost similar in different seasons.

Table 5. Weed composition, relative weed density (RWD) and relative weed biomass (RWB) in the untreated control plots in Boro 2014-15 at BRRI, Gazipur.

Weed species	Family	Class	RWD (%)	RWB (%)
Cynodon dactylon	Poaceae	Grass	10.76	9.21
Echinochloa crus-galli	Poaceae	Grass	31.48	33.76
Cyperus difformis	Cyperaceae	Sedge	32.50	34.07
Scirpus maritimus	Cyperaceae	Sedge	28.16	27.63
Monochoria vaginalis	Pontederiaceae	Broadleaf	21.42	26.74
Marsilea minuta	Marsileaceae	Broadleaf	10.90	12.65

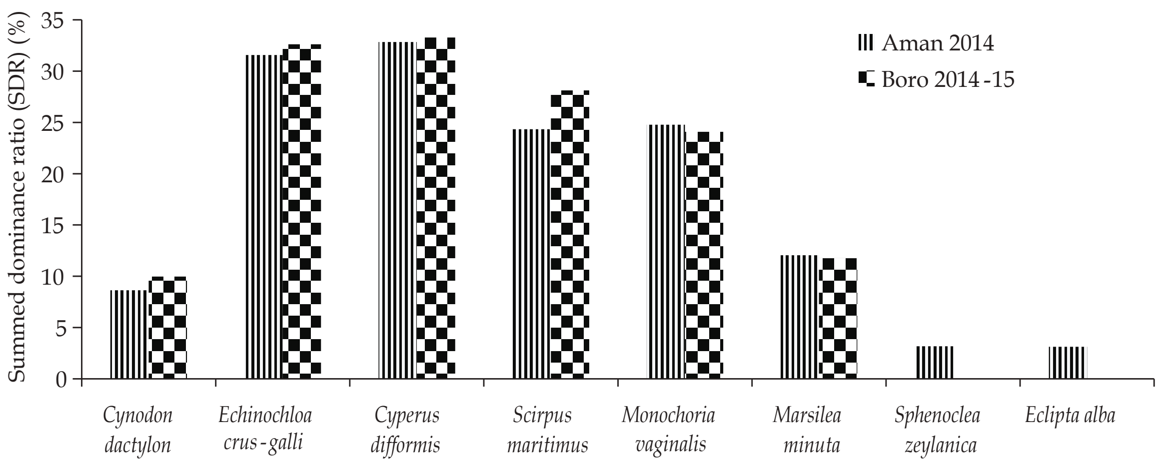


Fig. 1. Summed dominance ratio (SDR) of infesting weeds in transplanted rice.

Weed control efficiency (WCE)

Lower weed biomass as well as higher weed control efficiency was observed in all the growing seasons through Authority 48 SC. Weed control efficiency improved with the increases of herbicide dose irrespective of weed species. Treatment T_1 did not control all the weeds effectively (<80%) due to lower dose of application, whereas T_2, T_3 and T_4 control *Echinochloa crus-galli, Cyperus difformis, Scripus maritimus* and *Marsilea minuta* more than 80% in T. Aman season (Table 6). The trend of weed control efficiency in Boro 2014-15 was almost similar to T. Aman 2014 season. All the treatments controlled most of the weeds more than 80% except T_1. Treatments T_2, T_3 and T_4 controlled *Echinochloa crus-galli, Cyperus difformis, Scripus maritimus* and *Marsilea minuta* more than 80% (Table 7). It was evident from the study that pre planting herbicide Authority 48 SC @ 200 ml ha^{-1} and 250 ml ha^{-1} found effective for controlling weeds than other doses of that herbicide. Bhuiyan *et al*., 2017 reported that different groups of herbicides controlled more than 80% of weeds in different nitrogen management practices in hybrid rice.

Yield and yield attributes

Grain yield is the function of an interaction among various yield components, which were affected differentially by the growing conditions and crop management practices. All the treatments significantly increased rice grain yield over unweeded (control) plot (Table 8). In T. Aman 2014, the highest grain yield (5.22 t ha^{-1}) was recorded in T_2 treatment which was statistically similar to the treatments of T_4 and T_5 that produced 4.98 and 5.18 T ha^{-1} grain yield respectively. Minimum grain yield

Table 6. Effect of Authority 48 SC on weed control efficiency in transplanted rice in Aman 2014 at BRRI, Gazipur.

Weed	Weed control efficiency (%)			
	T_1	T_2	T_3	T_4
Cynodon dactylon	36.75	49.50	53.20	45.80
Echinochloa crus-galli	63.45	80.95	84.50	84.61
Cyperus difformis	70.25	82.60	83.65	81.54
Scripus maritimus	71.90	84.30	87.20	83.72
Monochoria vaginalis	65.45	74.65	78.70	80.59
Marsilea minuta	56.00	80.75	82.45	82.26
Sphenoclea zeylanica	52.40	73.55	78.90	72.65
Eclipta alba	54.20	63.50	66.90	62.75

T_1= Authority 48 SC @ 150 ml ha^{-1}, T_2= Authority 48 SC @ 200 ml ha^{-1}, T_3= Authority 48 SC @ 250 ml ha^{-1} and T_4= Pretilachlor @ 1000 ml ha^{-1}.

Table 7. Effect of Authority 48 SC on weed control efficiency in transplanted rice in Boro, 2014-15 at BRRI, Gazipur.

Weed	Weed control efficiency (%)			
	T_1	T_2	T_3	T_4
Cynodon dactylon	38.40	49.90	50.36	52.45
Echinochloa crus-galli	61.35	82.60	86.90	83.35
Cyperus difformis	72.40	82.15	85.60	85.60
Scripus maritimus	66.50	81.30	85.35	83.68
Monochoria vaginalis	60.05	75.80	78.20	83.40
Marsilea minuta	55.60	80.45	84.10	83.50

Table 8. Effect of Authority 48 SC on yield attributes of transplanted rice at BRRI, Gazipur.

Treatment	Panicle m^{-2}		Grain $panicle^{-1}$		Sterility (%)		TGW (g)		Grain yield (t ha^{-1})	
	S_1	S_2	S_1	S_2	S_1	S_2	S_1	S_2	S_1	S_2
T_1	226	261	17.70	16.87	105	112	21.86	25.41	4.87	5.34
T_2	235	286	16.13	14.63	107	116	21.87	25.44	5.22	5.67
T_3	214	255	18.10	17.67	96	104	21.75	25.42	4.69	4.97
T_4	237	283	17.27	15.27	103	114	21.76	25.53	4.98	5.69
T_5	236	287	16.23	14.60	107	117	21.93	25.27	5.18	5.75
T_6	186	214	19.77	19.07	78	93	20.84	24.92	3.21	3.37
CV (%)	4.56	4.03	4.05	9.06	3.47	5.14	2.2	1.1	3.48	3.51
LSD	18.48	19.35	1.29	2.69	6.28	10.26	ns	ns	0.29	0.32

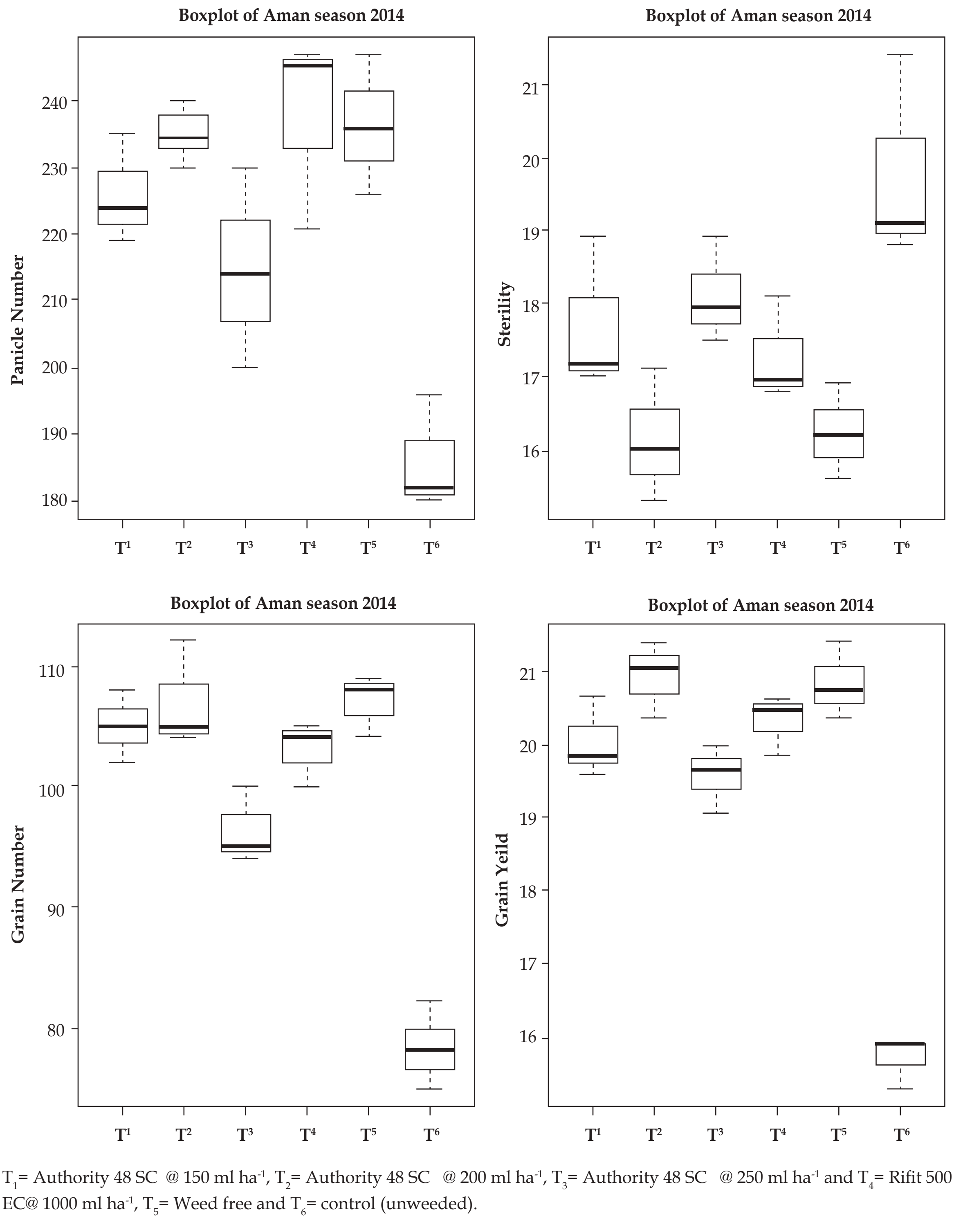
S_1= Aman 2014, S_2= Boro 2014-15. T_1= Authority 48 SC @ 150 ml ha^{-1}, T_2= Authority 48 SC @ 200 ml ha^{-1}, T_3= Authority 48 SC @ 250 ml ha^{-1} and T_4= Rifit 500 EC @ 1000 ml ha^{-1}, T_5= Weed free and T_6= control (unweeded).

(3.21 t ha^{-1}) was found in weedy check plots as compared to weed free treatment due to high weed density, which resulted less number of panicle m^{-2}, grains $panicle^{-1}$ and high sterility. Thousand grain weight (TGW) did not differ significantly among the treatments although unweeded plots produced lower TGW in both the years. Treatment wise boxplot of yield attributes in T. Aman 2014 confirmed that most of the yield contributing characters were similar to T_2, T_4 and T_5 (weed free) treatments; whereas T_6 was outsider of the normal range and its data was also in disperse condition than the other treatments due to severe weed infestation (Fig. 2).

Similar trend of results was observed during the Boro 2014-15 where unweeded control (T_6) produced minimum number of panicles m^{-2}, grains $panicle^{-1}$ and high sterility, which resulted lowest grain yield (3.37 t ha^{-1}). The minimum number of panicles m^{-2} in the control plot was probably due to the higher competition for nutrient, air space, light and water between crop plants and weeds which confirm the results of Hasanuzzaman *et al.*, 2009. Maximum grain yield of 5.75 t ha^{-1} was recorded with T_5 treatment due to lower weed-crop competition. In Boro 2014-15; T_2, T_4 and T_5 (weed free) treatments are in similar range in boxplot of yield attributes (Fig. 3). Authority 48 SC @ 150, 200, 250 ml ha^{-1} gave effective control of grass, sedge and broadleaf weeds lead to increased grain yield. Herbicide treatments contributed to higher yield performance compared to control in all the growing seasons (Bari, 2010).

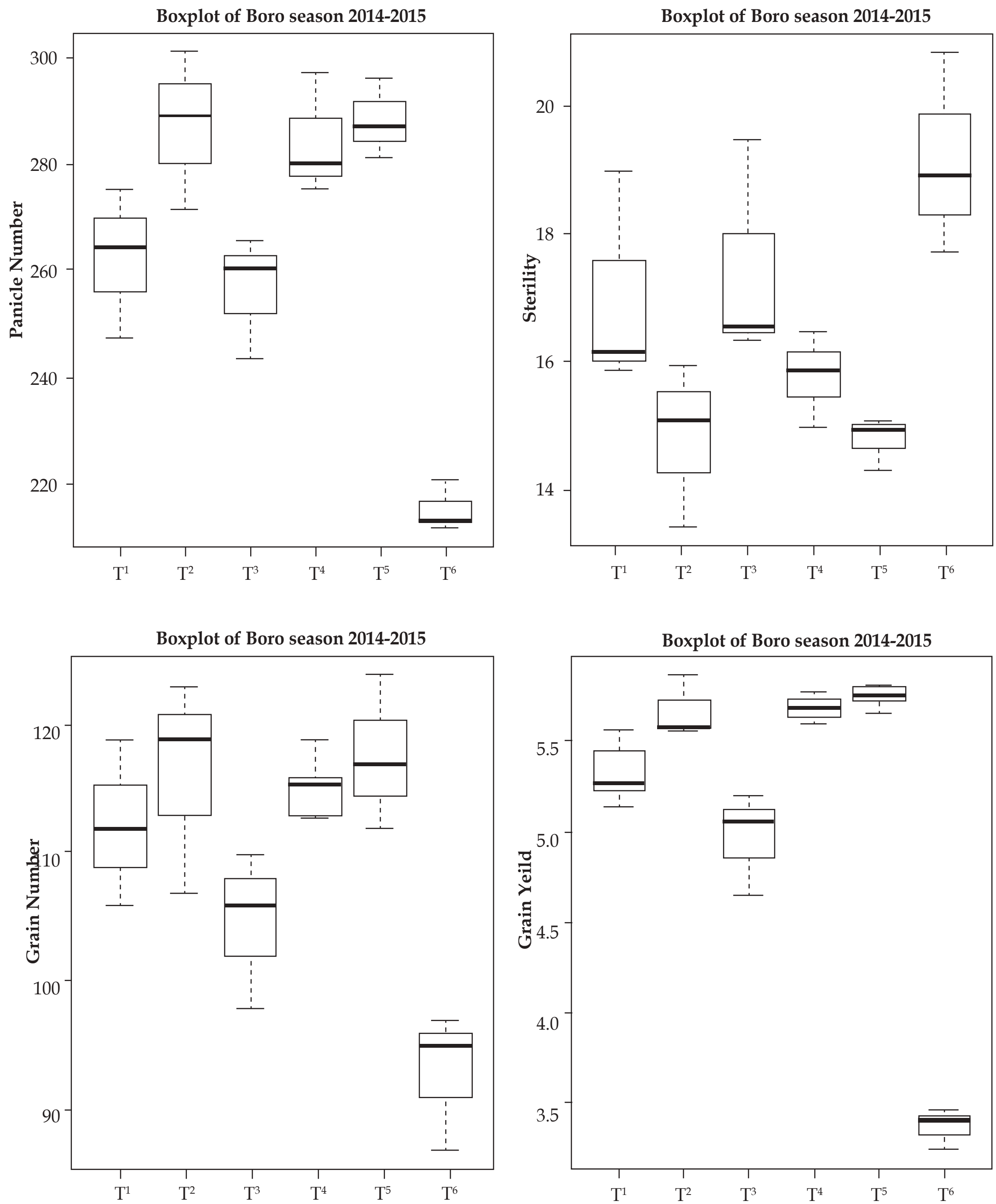
CONCLUSION

Grain yield and yield attributing parameters and weed dynamics were greatly influenced by different weed management practices. Authority 48 SC @ 250 ml ha^{-1} performed better weed control efficiency with slightly phytotoxity. Depending on the weeding efficacy, Authority 48 SC @ 200 ml ha^{-1} applied at three days before transplanting is effective for annual weed control option instead of hand weeding at peak period of labour crisis which can reduce the production cost.



T_1= Authority 48 SC @ 150 ml ha^{-1}, T_2= Authority 48 SC @ 200 ml ha^{-1}, T_3= Authority 48 SC @ 250 ml ha^{-1} and T_4= Rifit 500 EC@ 1000 ml ha^{-1}, T_5= Weed free and T_6= control (unweeded).

Fig. 2. Boxplot of yield attributes in Aman, 2014 at BRRI, Gazipur.



T_1= Authority 48 SC @ 150 ml ha^{-1}, T_2= Authority 48 SC @ 200 ml ha^{-1}, T_3= Authority 48 SC @ 250 ml ha^{-1} and T_4= Rifit 500EC @ 1000 ml ha^{-1}, T_5= Weed free and T_6= control (unweeded).

Fig. 3. Boxplot of yield attributes in Boro, 2014-15 at BRRI, Gazipur.

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Enhancing Rice Productivity through Integration of Stress Tolerant Rice Varieties and Improved Nutrient Management Practices in Saline Areas of Bangladesh

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ABSTRACT

The study was conducted in two locations of coastal districts Patuakhali and Satkhira during 2012 and 2013 T. Aman season. Stress tolerant rice varieties along with nitrogen application using prilled urea (PU), leaf color chart (LCC), urea super granule (USG), and rice crop manager (RCM) software based nitrogen (N) dose were examined. The objectives of the study were to identify the response of saline tolerant varieties to N fertilization on grain yield and profitability. Among the tested varieties, grain yield of BRRI dhan40, BRRI dhan41 and BRRI dhan54 were higher compared to BRRI dhan52 and BRRI dhan53 irrespective of location. There were no significant difference among the better performed varieties. Interaction effect of yield was significant in 2013 at Patuakhali but insignificant in both the locations in 2012. During 2013 in Patuakhali, the interaction effect of BRRI dhan40 × USG and BRRI dhan41 × USG produced higher grain yield and total N uptake. In Satkhira BRRI dhan54 and BRRI dhan40 performed better and produced higher grain yield and N uptake. Among the N application treatments USG application was the best compared to either LCC or RCM. The combination of BRRI dhan54×USG and BRRI dhan41×USG had more economic gains in both 2012 and 2013 in Patuakhali. The combination of BRRI dhan52×USG and BRRI dhan41×LCC appeared as themost profitable in Satkhira during 2013. Integration of saline tolerant varieties along with USG application could improve the yield of saline tolerant rice in saline environment.

Key words: Saline tolerant rice varieties, Saline soil, nitrogen management

INTRODUCTION

About 400-950 million hectares (mha) of land around the world (Lin *et al.*, 1998) is affected by different levels of salinity whereas about 0.83 mha land is affected by the salinity in Bangladesh (Karim *et al.*, 1990). In addition, the coastal areas share about 25% (2.5 mha) the total cropland in Bangladesh. Thus, soil salinity is the primary constraints toward the rice productivity in every rice growing country including Bangladesh. On the other hand, the imbalanced use of fertilizer and declining land productivity are the main concerns with the food insecurity in Bangladesh to feed the huge people (Uddin *et al*. (2011).

Salinity threatens both the plant root environment and hydrological situation and thereby devastating the normal crop production, but that varies across the crop seasons (Haque, 2006). The T. Aman rice is dominated by the low yield potential in Patuakhali. Notably, the average yield of T. Aman rice in Bangladesh is nearly 3.75 t ha^{-1} (AIS, 2015). The cropping pattern T. Aman-Fallow-Fallow is the most prevalent in the coastal areas of both Patuakhali and Satkhira. In fact, these areas were given minor attention in the past. Most recently, it is an imperative to explore the possibilities of the saline affected lands to increase the food grain productions.

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Salinity has a negligible impact due to the huge rainfall and upward flow of rivers when the crops are in the early phase of growth. In contrast, the soil salinity has an enormous effect on the later phase of crop due to the inadequacy of soil water at different soil profiles when the rain thoroughly stops. Moreover, soil salinity sometimes exceed the threshold limit of the tolerance of the rice crop. More certainly, salinity interrupts the plant growth and development and thereby causing the severe yield losses. Furthermore, the fertility level of most saline soils varies from low to very low according to the existence of the organic matter, nitrogen, phosphorus, and other micronutrients. That's why, proper doses of macro and micronutrients with the specific times are needed to meet up the nutrient requirements to boost up the crop productivity. It is also notable that traditional varieties, being very poor yielder are more commonly practiced in the saline areas. However, Bangladesh Rice Research Institute (BRRI) introduced the salt tolerant rice varieties. Among them, BRRI dhan40 and BRRI dhan41 are more popular in Aman season in the coastal region (CCC, 2009). They can easily survive in the salinity range of 2 to 6 dS m^{-1} soil electrical conductivity (EC) until there productive stage. More recently, BRRI dhan53 and BRRI dhan54, high yielder accompanied by better grain quality and shorter duration, were released to combat in the salinity. But they can tolerate more salinity ranging from 7 to 8 dS m^{-1} in the reproductive phase (BRKB, 2017). The high salinity tremendously affects the rice growth and yield but the proper soil and fertilizers management can ensure the better production (Aslam *et al*. (1989). To be certain, combination of the aforesaid modern varieties and the improved nutrient management were proposed to examine the potentials of rice productivity in the saline areas since the farmers in the saline area did not use the modern genotype as well as the recommended fertilizers, more specifically nitrogenous fertilizers. Considering the current situation in the saline areas, this piece of research was undertaken to examine the performance of newly released salt tolerant varieties in the farmers' field with different nitrogen fertilizer source and method of application.

MATERIALS AND METHODS

Experimental site characterization. The on-farm experiments were conducted at two coastal saline districts– Patukhali and Sathkira. In Patuakhali, Pakhimara village under Kalapara Thana (21°951829 N latitude and 90°3748354 E longitude, at an elevation of 0.65 m MSL), was selected and the experiments were conducted for the successive years during T. Aman 2012 and 2013. In 2013, an additional variety BRRI dhan52 and the nutrient recommendation from RCM of International Rice Research Institute were added. In Sathkira, Kulia village under Debhata Thana (22°6426074 N latitude and 88°9874502 E longitude with an altitude of 3.5 m from mean sea level), was also selected as an experimental site for the experiment only during T. Aman, 2013 (Table 1). The major soil type of Patuakhali is non-calcareous loam with the ranges from 1.7 to 3.4% organic content and slightly acidic having the pH 6.5-7.0. The available N, P, and K are 0.1-0.2%, 10-28 ppm, 0.2-0.6 meq%, respectively. The region is characterized by a close network of inter connected tidal rivers and creeks. The whole region of Patuakhali is lying within the cyclone zone and Satkhira lying in the Gangetic tidal floodplains. The main soil types are non-calcareous and clay loam having 1.8-2.2% organic matter and 6.2-8.4 pH. The available N, P and K are 0.1-0.3%, 12-24 ppm, 0.2-1.2 meq%, respectively. Maximum and minimum temperature, rainfall data were collected from the meteorological department and presented in (Fig. 1).

Experimental design and management. Table 1 presents the experimental treatments. The experiments were conducted in the split plot design where the varieties were used in the main plots and N-managements applied in the sub plots. In both the experimental locations during 2013, BRRI dhan52 and the crop nutrient

Table 1. Details of on-farm experiment conducted at Patuakhali in 2012 and 2013 and Sathkira in 2013 during T. Aman season.

Experimental site	Patuakhali	Patuakhali and Satkhira
Variety	BRRI dhan40	BRRI dhan40
	BRRI dhan41	BRRI dhan41
	BRRI dhan53	BRRI dhan52
	BRRI dhan54	BRRI dhan53
	-	BRRI dhan54
N management	PU	PU
	LCC	LCC
	USG	USG
	-	RCM
Fertilizer rate (kg ha^{-1})		
TSP-MoP-$ZnSO_4$. H_2O-gypsum	100-120-7.5-68	100-120-7.5-68
LCC-N	52	52
USG (1.8 g)-N	50	50
PU-N	69	69

TSP: Triple Super Phosphate, MoP: Muriate of Potash, PU: Prilled Urea, LCC: Leaf Colour Chart, USG: Urea Super Granule, RCM: Rice crop manager.

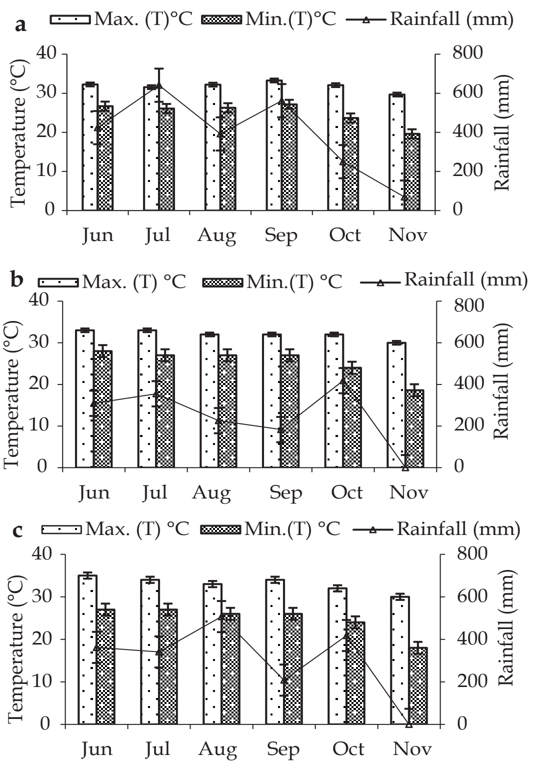


Fig. 1. Monthly average maximum and minimum temparature (°C) and total rainfall of Kalapara, Patuakhali district during (a) June 2012 to November 2012, (b) June 2013 to November 2013 and (c) Satkhira district during June 2013 to November 2013.

manager were also used. Tables 2 and 3 present details of crop nutrient manager. Nitrogen doses were recommended as 69, 52, and 50 kg ha^{-1} from PU, LCC, and USG respectively. The unit plot size was 5×6 m^2 with 20 ×15 cm row to row spacing and three farmers' plots were taken for three replications. BRRI recommended technology packages were followed in order to raise the seedlings and other intercultural operations. The fertilizer package 100-120-68-7.5 kg ha^{-1} that corresponds to triple super phosphate, muriate of potash, gypsum, and zinc sulphate were applied as basal following BRRI recommendation. In addition, PU was applied in three equal splits at 15 days after transplanting (DAT), maximum tillering and panicle initiation stage. LCC reading was recorded and urea was applied two times accordingly. USG (1.8g) was applied at 10 DAT in the middle of the alternate four hills. The 35-day-old seedlings was transplanted on 4th August 2012 in Patuakhali, whereas transplanted on 1st and 6th August, 2013 in Patuakhali and Satkhira site respectively. Simultaneously, water salinity was recorded at every seven days interval. The standard recommended plant protection measures were adopted to ensure the uninterrupted crop growth.

Table 2. Details of on-farm RCM treatment conducted at Kalapara, Patuakhali during T. Aman 2013.

Variety and growth stage	Date of application	Fertilizer for 33 decimals (kg)		
		TSP	MOP	Urea
BRRI dhan40				
Basal	0	4	2	-
Early	Aug 14-18	-	-	7
Active tillering	Sept 6-10	-	-	8
Panicle initiation	Sept 22-26	-	-	8
BRRI dhan41				
Basal	0	4	2	-
Early	Aug 14-18	-	-	7
Active tillering	Sept 11-15	-	-	8
Panicle initiation	Sept 27-31	-	-	8
BRRI dhan52				
Basal	0	4	2	-
Early	Aug 14-18	-	-	7
Active tillering	Sept 6-10	-	-	8
Panicle initiation	Sept 22-26	-	-	8
BRRI dhan53				
Basal	0	3	4	-
Early	Aug 14-18	-	-	7
Active tillering	Aug 24-28	-	-	-
Panicle initiation	Sept 2-6	-	-	10
BRRI dhan54				
Basal	0	4	2	-
Early	Aug 14-18	-	-	7
Active tillering	Aug 27-31	-	-	8
Panicle initiation	Sept 12-16	-	-	8

Data collection and statistical analysis. Data on yield and yield character were calculated according to Gomez K A (1972). Straw and grain samples were stored for N content estimation. The samples were oven dried at 70°C for 72 hours, weighed, ground, and then subsamples were taken for N determination. The N content in straw and grains was measured by the standard micro-Kjeldahl procedure (Bremner and Mulvaney, 1982). The N-uptake in grain and straw was calculated by following formulae.

Finally, the collected data were analyzed with software CROPSTAT 7.2. The least significant difference (LSD) at 5% probability was used to compare the means of the treatments (Gomez and Gomez, 1984).

Economic analysis. Economic comparison of the treatments were checked based on the production cost, gross return, net return, and benefit-cost ratio (BCR). The total variable cost was calculated by the inputcosts (seeds, fertilizers and pesticides); costs of human labour for land preparation, irrigation,

$$\text{Nitrogen uptake by grain (kg ha}^{-1}\text{)} = \frac{\text{\% N in grain} \times \text{grain yield (kg ha}^{-1}\text{)}}{100}$$

$$\text{Nitrogen uptake by straw (kg ha}^{-1}\text{)} = \frac{\text{\% N in straw} \times \text{straw yield (kg ha}^{-1}\text{)}}{100}$$

Table 3. Details of on-farm RCM treatment conducted at Debhata, Satkhira during T. Aman 2013.

Variety and growth stage	Date of application	Fertilizer for 33 decimals (kg)			
		TSP	MOP	Urea	Zinc sulphate
	BRRI dhan40				
Basal	0	4	2	0	0.7
Early	Aug 14-18	-	-	7	-
Active tillering	Sept 6-10	-	-	8	-
Panicle initiation	Sept 22-26	-	-	8	-
	BRRI dhan41				
Basal	0	4	2	-	0.7
Early	Aug 14-18	-	-	7	-
Active tillering	Sept 11-15	-	-	8	-
Panicle initiation	Sept 27-31	-	-	8	-
	BRRI dhan52				
Basal	0	4	2	-	0.7
Early	Aug 14-18	-	-	7	-
Active tillering	Sept 16-20	-	-	8	-
Panicle initiation	Oct 1-5	-	-	8	-
	BRRI dhan53				
Basal	0	4	4	-	-
Early	Aug14-18	-	-	9	-
Active tillering	Aug 24-27	-	-		-
Panicle initiation	Sept 2-6	-	-	12	-
	BRRI dhan54				
Basal	0	4	2	-	0.7
Early	Aug 14-18	-	-	7	-
Active tillering	Aug 27-31	-	-	8	-
Panicle initiation	Sept 12-16	-	-	8	-

fertilizer, pesticide applications, harvesting, bundling, carrying, and threshingthe rent of a power tiller and irrigation cost. Gross return was calculated by multiplying the quantity of production (grain and straw) by the output price at the harvest time. The net return and BCR were computed as follows:

Net return = Gross return-cost of production,

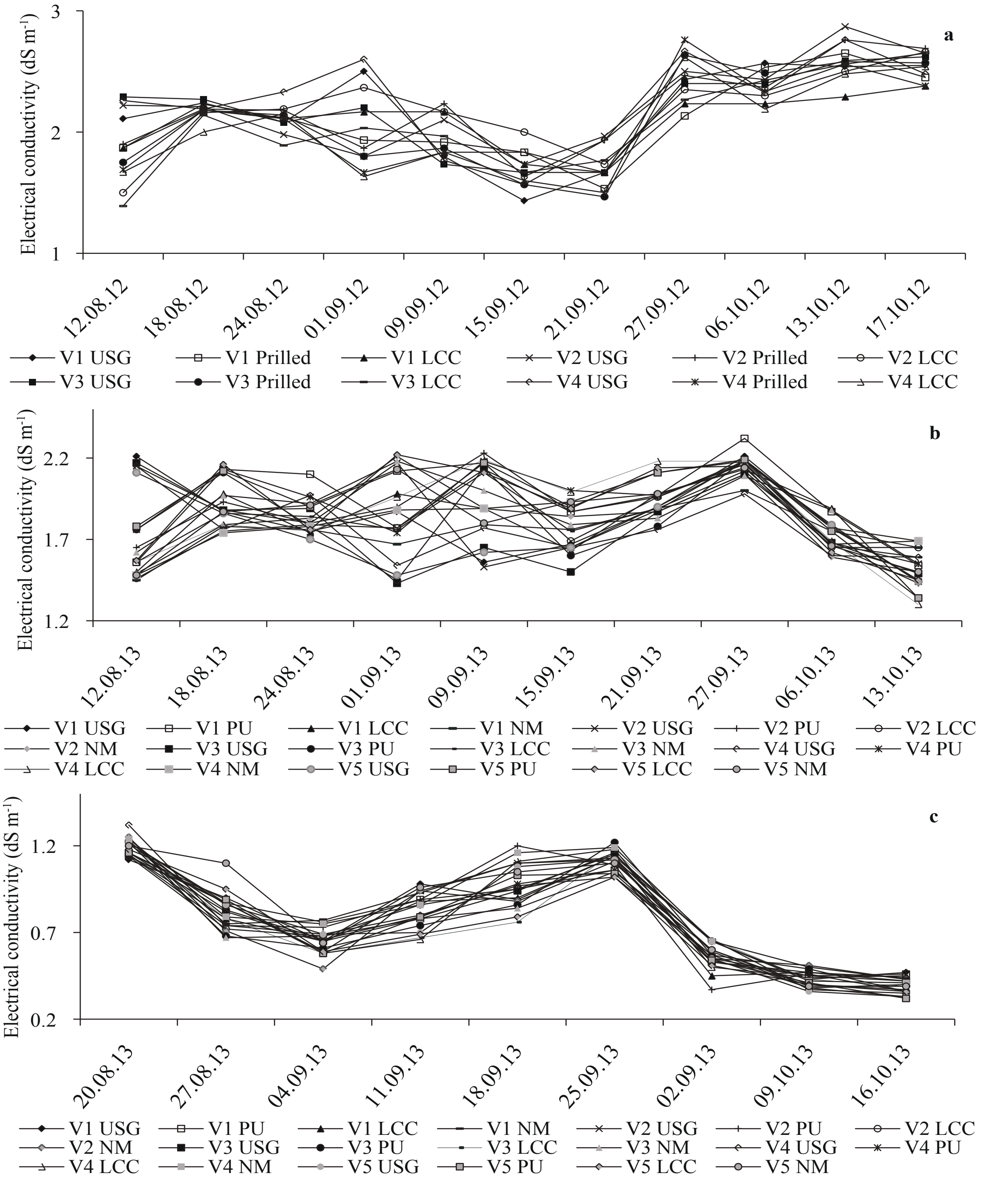
BCR = Gross return / cost of production.

The economic analysis was conducted by taking into account the prevailing market price of inputs, labours and produce during the year 2012-13 in Bangladeshi Taka (BDT) and then converted into US$ using the conversion rate 1US$ = 78BDT.

RESULTS AND DISCUSSION

Effect of water salinity. The on–farm field crop result showed that all the newly developed saline tolerant varieties grew very well as there wasa little salinity impact on crop production due to the better rainfall throughout the experiment period (Fig. 1). In both Pakhimara of Patuakhali, and Kulia of Satkhira, the fluctuation of soil salinity throughout the on-farm crop growing season was documented at weekly interval (Fig. 2). In Patuakhali during 2012 and 2013, water salinity varied from 1.39 to 2.87 and 1.30 to 2.32 dS m^{-1} respectively. In Satkhira, water salinity varied from 0.32 to 1.32 dS m^{-1} in the experimental plot for the same season. In Patuakali's on-farm experimental site, the salinity level was higher than that of Satkhira. However, the low intent of soil salinity did not cause any detrimental effect on the crop development and productivity atboth the experimental sites.

Yield attributing characters, yield and nitrogen uptake at Patuakhali in 2012 and 2013. Table 4 presents plant height at maturity, yield attributes and grain yield during 2012 at Patuakhali. Crop performance showed that in



V1: BRRI dhan40, V2: BRRI dhan41, V3: BRRI dhan53, V4: BRRI dhan54, V5: BRRI dhan52; PU: Prilled urea, LCC: Leaf colour chart, USG: Urea super granule, NM: Nutrient manager (RCM) V5 and NM was introduced during 2013 at Patuakhali and Satkhira.

Fig. 2. Weekly fluctuation of salinity at farmers' field a). and b). Pakhimara, Patuakhali during T. Aman 2012 and 2013, respectively; and c). Kulia, Satkhira during T. Aman 2013.

Table 4. Plant height, yield attributing characters and grain yield in different salt tolerant varieties, influenced by N management at Kalapara, Patuakhali during T. Aman 2012.

Treatment	Plant height (cm)	Panicle m^{-2}	Grain $panicle^{-1}$	1000 grain wt (g)	Panicle length (cm)	Grain yield (t ha^{-1})
			Variety			
BRRI dhan40	109.6	196.00	78	24.72	24.93	4.16
BRRI dhan41	113.14	191.67	74	25.31	25.27	4.09
BRRI dhan53	108.25	164.22	63	23.53	21.68	3.57
BRRI dhan54	102.11	185.44	81	25.32	24.48	4.29
$LSD_{0.05}$	5.64	^{a}ns	10.6	0.68	1.19	0.40
			N management			
USG	107.87	193.53	78	25.01	24.37	4.22
PU	108.41	180.58	72	24.53	23.83	3.94
LCC	108.57	178.83	72	24.62	24.08	3.92
$LSD_{0.05}$	ns	ns	ns	ns	ns	ns

ns: not significant, PU: Prilled urea, LCC: Leaf colour chart, USG: Urea super granule.

the main plot, plant height at harvest of different varieties differed significantly whereas nitrogen management had no significant influence on the plant height of varieties. There was no significant effect of panicle m^{-2} in main plot and sub plots, although in main plot, BRRI dhan40 produced higher panicles m^{-2} (196) and in sub plot USG treated plots produced higher panicles m^{-2} (193). Grains $panicle^{-1}$, 1000 grain weight (TGW) and panicle length of the varieties differed significantly in the main plot but significant difference in sub plot (N management). The highest grains $panicle^{-1}$ in main plot was observed in BRRI dhan40 closely followed by BRRI dhan54 and the lowest was produced from BRRI dhan53. Grain weight (1000-seed) was the highest in BRRI dhan54 followed by BRRI dhan41 and the lowest was observed from BRRI dhan53. Length of panicle varied significantly in both varieties and nitrogen management. Regarding main effects of varieties, the highest grain yield was observed in BRRI dhan54 (4.29 t ha^{-1}) and the lowest yield (3.57 t ha^{-1}) in BRRI dhan53. Regarding N management treatment, USG produced the highest grain yield (4.22 t ha^{-1}) followed by prilled urea (3.94 t ha^{-1}). The lowest grain yield (3.92t ha^{-1}) was recorded from LCC treatment.

Table 5 presents varietal effect and nitrogen management on nitrogen uptake in grain and straw. Varieties and nitrogen management differed significantly on grain and straw nitrogen uptake. In the main plot, nitrogen uptake in grain (25.96 kg ha^{-1}) and straw (11.40 kg ha^{-1}) was found the highest in BRRI dhan41 and in sub plot N applied through USG (27.43 kg ha^{-1} in grain and 11.81 kg ha^{-1} in straw). The lowest nitrogen uptake, in grain and straw, was found in BRRI dhan53 and N management through LCC treatment. Grain yield was found significantly and linearly related with total N uptake (Fig. 3a) indicating that higher grain yield would be due to higher N uptake. Kabir *et al.* (2011) observed variation in N uptake by grain was 25.14 to 48.02 kg ha^{-1} at Satkhira district, Bangladesh in STL-655 rice mutant cultivar. Similarly, the range of N uptake by straw was 20.36 to 35.85 kg ha^{-1}.

Table 6 presents plant height at maturity, yield and yield attributing characters. In 2013 at Patuakhali crop performance showed that plant height at harvest differed significantly in main plot, but in nitrogen management plot (sub plot) did not influenced significantly. There were no significant effect on panicle m^{-2}, grain $panicle^{-1}$ and TGW of the varieties in main plot; but in sub plot, nitrogen management significantly affected on panicle m^{-2}, grain $panicle^{-1}$ and TGW. Length of panicles varied significantly

Table 5. Nitrogen uptake in grain and straw of different salt tolerant varieties, influenced by N management at Kalapara, Patuakhali during T. Aman 2012.

Treatment	N uptake (kg ha^{-1})	
	Grain	Straw
Variety		
BRRI dhan40	25.96	11.40
BRRI dhan41	24.77	10.77
BRRI dhan53	21.41	8.75
BRRI dhan54	27.04	11.91
$LSD_{0.05}$	2.66	1.64
N management		
USG	27.43	11.81
PU	23.51	10.08
LCC	23.45	10.24
$LSD_{0.05}$	1.90	1.01

PU: Prilled urea, LCC: Leaf colour chart, USG: Urea super granule.

both in varieties and nitrogen management. Regarding main effects of varieties, the highest grain yield was recorded in BRRI dhan40 (4.53 t ha^{-1}) and the lowest yield (2.88 t ha^{-1}) was recorded in BRRI dhan53. Regarding nitrogen management treatment, USG produced the highest grain yield (5.03 t ha^{-1}) followed by

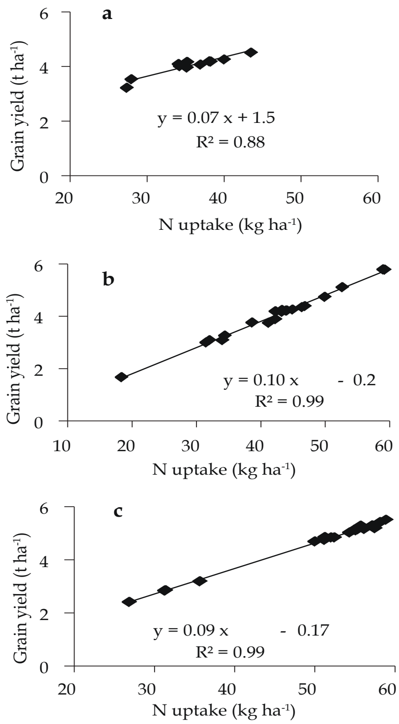


Fig. 3. Relationship among grain yield and N uptake in salt tolerant varieties at a). and b). Patuakhali during T. Aman 2012 and 2013 respectively; and c) Satkhira during T. Aman 2013.

Table 6. Plant height, yield and yield attributing characters of different salt tolerant varieties, influenced by N management at Kalapara, Patuakhali during T. Aman 2013.

Treatment	Plant height (cm)	Panicle m^{-2}	Grains panicle^{-1}	1000 grain wt (g)	Panicle length (cm)	Grain yield (t ha^{-1})
Variety						
BRRI dhan40	112.35	176.08	71	25.98	25.86	4.53
BRRI dhan41	115.91	172.83	77	25.86	26.31	4.36
BRRI dhan53	104.80	136.58	62	26.02	21.28	2.88
BRRI dhan54	100.58	164.83	71	25.57	21.68	4.30
BRRI dhan52	108.15	164.58	71	25.65	21.17	4.17
$LSD_{0.05}$	7.64	ns	ns	ns	2.21	0.74
N management						
USG	107.90	175.93	81.35	27.03	24.89	5.03
PU	109.26	167.93	71.47	25.92	23.18	4.04
LCC	109.37	160.80	71.22	25.30	22.80	3.98
RCM	106.93	147.26	58.08	25.01	22.17	3.13
$LSD_{0.05}$	ns	12.43	5.80	0.98	1.23	0.28

ns: not significant, PU: Prilled urea, LCC: Leaf colour chart, USG: Urea super granule, RCM: Rice crop manager.

prilled urea (4.04 t ha^{-1}). The lowest grain yield (3.13 t ha^{-1}) was observed in nutrient manager treatment. Interaction effect of varieties and nitrogen management differed significantly on grain yield and nitrogen uptake (Table 7). BRRI dhan41 × USG produced the highest grain yield (5.79 t ha^{-1}) which was statistically similar with the BRRI dhan40 × USG (5.78 t ha^{-1}). The lowest grain yield (1.67 t ha^{-1}) was recorded from BRRI dhan53 × nutrient manager treatment. The highest nitrogen uptake in grain was observed in BRRI dhan41 × USG treatment (59.21 kg ha^{-1}) followed by BRRI dhan40 × USG (58.96 kg ha^{-1}). The lowest nitrogen uptake in grain was from BRRI dhan53 × nutrient manager treatment (18.37 kg ha^{-1}). Nitrogen uptake in grain + straw was highest in BRRI dhan40 × USG followed by BRRI dhan41 × USG treatment. Rice varieties varied in yield responses to applied nitrogen, have been reported by Fageria and Barbosa Filho (2001), Fageria and Baligar (2005). Similarly, genotypic variations in N uptake and utilization have been reported by Fageria and Baligar, (2005).

Table 7. Interaction effect of salt tolerant varieties and N management on grain yield and grain N uptake at Kalapara, Patuakhali during T. Aman 2013.

N management (NM)		Grain yield (t ha^{-1})	Grain N uptake (kg ha^{-1})
	BRRI dhan40		
USG		5.78	41.87
PU		4.39	31.90
LCC		4.19	29.92
RCM		3.75	27.95
	BRRI dhan41		
USG		5.79	42.67
PU		4.24	30.83
LCC		4.17	30.13
RCM		3.26	23.71
	BRRI dhan53		
USG		3.76	26.99
PU		3.00	21.62
LCC		3.09	22.28
RCM		1.67	12.33
	BRRI dhan54		
USG		4.74	34.16
PU		4.35	31.95
LCC		4.22	30.12
RCM		3.89	29.31
	BRRI dhan52		
USG		5.11	36.72
PU		4.22	30.93
LCC		4.26	30.87
RCM		3.09	22.94
$LSD_{0.05}$	V	0.74	5.27
RCM		0.28	2.09
V × RCM		0.62	4.67

PU: prilled urea, LCC: Leaf colour chart, USG: Urea super granule, RCM: Rcie crop manager.

Yield attributing characters, yield and nitrogen uptake at Satkhira in 2013. Table 8 presents plant height at maturity, yield and yield attributes of rice varieties, and nitrogen management at Satkhira district during 2013. Varieties differed significantly regarding plant height, panicle m^{-2}, panicle length and grain yield. TGW was not differed significantly in main plot. The highest plant height was observed in BRRI dhan41 and the lowest was in BRRI dhan54. The highest panicles m^{-2} (219) was produced from BRRI dhan54 followed by BRRI dhan40. The lowest panicle was produced in BRRI dhan53. The highest grains panicle^{-1} (86) was observed in BRRI dhan54 followed by BRRI dhan40 (85) and the lowest (63) was observed in BRRI dhan53. The highest panicle length (cm) was observed in BRRI dhan41 (26.15 cm) followed by BRRI dhan40 (26.10 cm). The lowest panicle length was recorded from BRRI dhan52 (22.48). Higher yield attributing characters lead to achieve highest grain yield in BRRI dhan54 (5.22 t ha^{-1}) followed by BRRI dhan41 (5.18 t ha^{-1}) (Table 8). Nitrogen management differed significantly on yield attributing characters. The highest panicles m^{-2}, grains panicle^{-1}, panicle length and TGW were observed in USG treatment irrespective of variety, which lead to produce highest grain yield (4.96 t ha^{-1}) followed by prilled urea. The lowest grain yield was produced from nutrient manager treatment (4.41 t ha^{-1}). N uptake in grain and straw differed significantly (Table 9). The highest N uptake in grain was recorded in BRRI dhan40 followed by BRRI dhan54 and the lowest in BRRI dhan53. Nitrogen uptake in grain and straw due to nitrogen management were not differed significantly. Islam *et al.* (2011) observed that in T. Aman season, plant

Table 8. Plant height and yield attributing characters of different salt tolerant varieties, influenced by N management at Debhata, Satkhira during T. Aman 2013.

Treatment	Plant height (cm)	Panicle m^{-2}	Grain $panicle^{-1}$	1000 grain wt (g)	Panicle length (cm)	Grain yield (t ha^{-1})
			Variety			
BRRI dhan40	113.29	218.33	85.33	26.34	26.10	5.18
BRRI dhan41	116.00	207.41	79.16	26.67	26.15	4.97
BRRI dhan53	108.33	176.33	63.08	24.95	23.29	2.82
BRRI dhan54	104.32	219.75	85.83	24.72	23.38	5.22
BRRI dhan52	108.77	217.16	77.58	24.92	22.48	5.03
$LSD_{0.05}$	5.85	21.69	6.16	ns	0.96	*0.26*
			N management			
USG	110.38	220.13	84.20	26.29	25.38	4.96
PU	110.73	209.93	79.53	25.65	24.57	4.65
LCC	110.71	206.20	75.73	25.25	23.96	4.57
RCM	108.75	194.93	73.33	24.88	23.21	4.41
$LSD_{0.05}$	ns	11.89	4.58	0.75	1.17	*0.32*

PU: Prilled urea, LCC: Leaf colour chart, USG: Urea super granule, RCM: Rice crop manager, ns: not significant.

Table 9. Nitrogen uptake in grain and straw of different salt tolerant varieties, influenced by N management at Debhata, Satkhira during T. Aman 2013.

Treatment	N uptake (kg ha^{-1})	
	Grain	Straw
	Variety	
BRRI dhan40	40.30	15.65
BRRI dhan41	37.82	15.23
BRRI dhan53	21.65	9.66
BRRI dhan54	39.06	17.28
BRRI dhan52	37.65	16.57
$LSD_{0.05}$	2.84	1.35
	N management	
USG	37.46	15.82
PU	35.26	14.94
LCC	35.21	14.53
RCM	33.25	14.23
$LSD_{0.05}$	ns	ns

PU: Prilled urea, LCC: Leaf colour chart, USG: Urea super granule, RCM: Rice crop manager, ns: not significant.

height, number of tillers, total dry matter (TDM), length of panicles, number of filled grains, TGW and grain yield were gradually decreased with the increase level of salinity. Sharma *et al.*, 2013 recommended that BRRI dhan40, BRRI dhan41, BRRI dhan51, BRRI dhan52, BRRI dhan53 and BRRI dhan54 are suitable for cultivation in T. Aman season to improve the productivity of southern coastal region of Bangladesh where salinity level are low. Genotypic variation in grain yield, straw yield and nitrogen uptake by grain and straw were also reported by Saleque *et al*. (2004).

Economic analysis. The production cost was calculated based on the prices in local market of Patuakali and Satkhira during T. Aman, 2012 and 2013. A total of US$ 660 and 703 ha^{-1} was an average cost of rice production during 2012 and 2013 in Patuakhali and Satkhira respectively. Treatment dependent costs of cultivation were calculated on the basis of the additional inputs needed (Tables 10, 11 and 12). The BRRI dhan54 × USG showed the highest benefit cost ratio (BCR; 1.78) (Table 10). The calculated BCR showed that all treatment combinations had a BCR above 1.5 except BRRI dhan54 × LCC (1.29) and BRRI dhan53 × LCC (1.36). A BCR more than 2 appears to be good investment that yields a double return per unit investment (Reddy and Reddy,

Table 10. Economic analyses (US $ ha^{-1}) of cost and return of N management with new salt tolerant varieties in on-farm studies at Kalapara, Patuakhali during T. Aman 2012.

Treatment	Cost of cultivation	Gross return	Net return	BCR
BRRI dhan40 × PU	724	1144	421	1.58
BRRI dhan41 × PU	724	1163	440	1.61
BRRI dhan53 × PU	724	1168	444	1.61
BRRI dhan54 × PU	724	1124	400	1.55
BRRI dhan40 × LCC	722	1147	425	1.59
BRRI dhan41 × LCC	722	1163	441	1.61
BRRI dhan53 × LCC	722	982	261	1.36
BRRI dhan54 × LCC	722	929	207	1.29
BRRI dhan40 × USG	705	1117	412	1.58
BRRI dhan41 × USG	705	1160	454	1.64
BRRI dhan53 × USG	705	1168	463	1.66
BRRI dhan54 × USG	705	1257	552	1.78

PU: Prilled urea, LCC: Leaf colour chart, USG: Urea super granule.

Table 11. Economic analyses (US $ ha^{-1}) of cost and return of N management with new salt tolerant varieties in on-farm studies at Kalapara, Patuakhali during T. Aman 2013.

Treatment	Cost of cultivation	Gross return	Net return	BCR
BRRI dhan40 × PU	766	1227	461	1.60
BRRI dhan41 × PU	766	1187	421	1.55
BRRI dhan53 × PU	766	841	75	1.10
BRRI dhan54 × PU	766	1229	463	1.60
BRRI dhan52 × PU	766	1183	417	1.54
BRRI dhan40 × LCC	764	1172	408	1.53
BRRI dhan41 × LCC	764	1170	406	1.53
BRRI dhan53 × LCC	764	866	102	1.13
BRRI dhan54 × LCC	764	1191	426	1.56
BRRI dhan52 × LCC	764	1197	432	1.57
BRRI dhan40 × USG	748	1610	862	2.15
BRRI dhan41 × USG	748	1612	865	2.16
BRRI dhan53 × USG	748	1054	307	1.41
BRRI dhan54 × USG	748	1328	581	1.78
BRRI dhan52 × USG	748	1429	682	1.91
BRRI dhan40 × RCM	771	1063	291	1.38
BRRI dhan41 × RCM	771	916	145	1.19
BRRI dhan53 × RCM	748	472	-276	0.63
BRRI dhan54 × RCM	771	1094	323	1.42
BRRI dhan52 × RCM	771	871	100	1.13

PU: Prilled urea, LCC: Leaf colour chart, USG: Urea super granule, RCM: Rice crop manager.

Table 12. Economic analyses (US $ ha^{-1}) of cost and return of N management with new salt tolerant varieties in on-farm studies at Debhat, Satkhira during T. Aman 2013.

Treatment	Cost of cultivation	Gross return	Net return	BCR
BRRI dhan40 × PU	749	1437	687	1.92
BRRI dhan41 × PU	749	1436	687	1.92
BRRI dhan53 × PU	749	1452	703	1.94
BRRI dhan54 × PU	749	1415	665	1.89
BRRI dhan52 × PU	749	1373	623	1.83
BRRI dhan40 × LCC	760	1348	588	1.77
BRRI dhan41 × LCC	760	1520	760	2.00
BRRI dhan53 × LCC	760	1269	509	1.67
BRRI dhan54 × LCC	760	849	90	1.12
BRRI dhan52 × LCC	760	849	89	1.12
BRRI dhan40 × USG	731	923	192	1.26
BRRI dhan41 × USG	731	725	-6	0.99
BRRI dhan53 × USG	731	1424	693	1.95
BRRI dhan54 × USG	731	1450	719	1.98
BRRI dhan52 × USG	731	1467	736	2.01
BRRI dhan40 × RCM	761	1422	661	1.87
BRRI dhan41 × RCM	761	1420	659	1.87
BRRI dhan53 × RCM	739	1298	560	1.76
BRRI dhan54 × RCM	761	1511	750	1.99
BRRI dhan52 × RCM	761	1333	572	1.75

PU: Prilled urea, LCC: Leaf colour chart, USG: Urea super granule, NM: Nutrient manager.

1992). Moreover, a BCR at least above 1.5 has been considered economically viable for an agricultural enterprise (Makarim *et al*., 2002). Thus, the calculated BCR, (Table 10) indicate that all tested treatments except BRRI dhan54 × LCC and BRRI dhan53 × LCC were economically viable. The actual net returns showed that some treatments were much more profitable than the others. In this respects, treatment BRRI dhan54 × USG showed the higher net return of US$ 552 ha^{-1} in 2012 at Patuakhali. In Patuakhali during 2013, the higher benefit were observed in the treatments BRRI dhan40 × USG (2.15) and BRRI dhan41 × USG (2.16) and lower benefits were observed in the treatments BRRI dhan53 × prilled urea (1.10), BRRI dhan53 × LCC (1.13), BRRI dhan53 × nutrient manager (0.63), BRRI dhan41 × nutrient manager (1.19) and BRRI dhan54 × nutrient manager (1.13). In Satkhira, the higher benefits were observed in the treatments of BRRI dhan52 × USG (2.01), BRRI dhan41 × LCC (2.00). The lower benefits were found with the treatments of BRRI dhan41 × USG (0.99), BRRI dhan54 × LCC (1.12) and BRRI dhan52 × LCC (1.12) treatments. The experimental results clearly indicate that the application of USG gave more return than PU, LCC, and nutrient manager.

CONCLUSION

This study showed that nitrogen application through USG enhanced the growth, yield and yield attributes of rice as well as nitrogen uptake in both grain and straw. Among all the tested varieties, BRRI dhan40, BRRI dhan41 and BRRI dhan54 was better in both the locations. USG application was performed better in both the location and year. Rice crop manager needs to be updated for saline ecosystem. Improving nitrogen fertilization in coastal saline rice culture has potential to increase grain yield of saline tolerant rice. Such a cost effective and promising technology for the stress tolerant rice varieties can enhance the yield potential as well as upgrade the livelihood of the poor farmers in the study regions.

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Recovering Ability of Upland and Rainfed Lowland Rice Varieties against Rice Tungro Disease

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ABSTRACT

Rice tungro is the most destructive and widespread among virus diseases found in almost all rice growing areas. In susceptible varieties, it causes 100% yield loss under favourable environmental conditions. Control of tungro disease by chemical applications is not effective and eco-friendly. Development of resistant variety against tungro is also difficult, because it is necessary to develop resistance either to the insect vector or to the virus or to the both. Identification of varieties with recovering ability will facilitate farmers to cultivate rice in tungro endemic areas. Bangladesh Rice Research Institute released six upland and eight rainfed lowland rice varieties, which were tested to evaluate their recovering ability against tungro disease. The test varieties including susceptible and resistant checks were artificially inoculated by viruliferous vector (Green leafhopper, *Nephotettix virescens*) of ten days after seeding in net house condition. Among the tested varieties BR26, BRRI dhan33 and BRRI dhan40 were the most susceptible and showed high infection rate against tungro disease. BRRI dhan37 and BR8 showed the highest recovering ability in rainfed lowland and upland rice, respectively, with the lowest yield reduction compared to the resistant check Kumragoir, which could be used in tungro endemic areas. From these investigations, all the tested varieties expressed distinct tungro disease symptoms after three weeks of inoculation. With the advancement of plant growth, varieties BR8 and BRRI dhan37 recovered from tungro syndrome due to their genetic makeup consisting tolerance potentiality showed better yield, while other varieties like BRRI dhan33 and BRRI dhan40 did not produce any grain yield.

Key words: Tolerance, Aus, Aman, rice tungro virus

INTRODUCTION

Rice (*Oryza sativa* L.) is the most used cereal grain in the diet of more than three billion people in the world. Seventy-two diseases affect rice (Bergonia, 1978) and 22 of them are caused by virus and virus like microorganisms of which rice tungro is the most important (Ou, 1984). It is the most serious and wide spread disease occurring in the rice growing countries like Bangladesh (Miah, 1973), India (John, 1968), Malaysia (Ou *et al.*, 1965) and the Philippines (Rivera and Ou, 1965). Rice tungro disease (RTD), caused by the co-infection of rice tungro bacilliform virus (RTBV) and rice tungro spherical virus (RTSV). Both RTBV and RTSV are transmitted in a semi persistent manner by the green leafhopper (GLH), *Nephotettix virescens* (Distant), and some other leafhopper species (Hibino *et al.,* 1979; Hibino, 1983). For a susceptible variety without any recovery ability, rice tungro disease (RTD) may cause 100% infection and resulting in total yield loss under favourable condition (BRRI, 1983). The disease remains one of the major threat to sustainable rice production in many rice growing countries. It is one of the major constraints in rice production, particularly in the upland and rainfed ecosystems (Miah, 1984; Latif *et al.,* 2011). The lack of resistant genes to RTBV- the causal agent of tungro-disease makes it even more difficult to manage RTD (Dahal *et*

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al., 1992). Therefore, sustainable strategies are urgently required for the management of RTD.

The use of resistant or varieties with recovering ability is believed to be the most effective method for controlling rice tungro disease as killing the vector with insecticides to control this disease is not very effective and may cause environmental and health hazards (Latif *et al.,* 2011). Several rice germplasm sources resistant or tolerant to RTD have been used to improve rice cultivars in endemic areas (Khush *et al.,* 2004), although a distinction between resistance to GLH and to tungro viruses in some rice genotypes is still unclear (Shibata *et al.,* 2007; Zenna *et al.,* 2008). Some traditional rice cultivars and wild *Oryza* spp. have been used for the improvement of cultivars tolerant or resistant to RTD. Advanced breeding lines derived from Utri Merah consistently showed low infections with both RTBV and RTSV in several field trials (Cabunagan *et al.,* 1999). Kachamota and ARC 11554 were found resistant to tungro in Malaysia and Indonesia. Moreover, ARC 11554 was found resistant to tungro in many countries (Miah, 1984). Due to evolution of new strains, varieties of high recovering ability may also lose its potentiality. Therefore, it is pertinent to evaluate existing rice varieties having recovering ability, which are intensively cultivated in the farmer's field. Present studies were conducted to identify the varieties, which have high recovering ability against tungro disease and these might be used to cultivate under rainfed upland and lowland ecosystems in tungro endemic areas. The varieties which recovered from disease will also be used as parents for hybridization programme to develop tungro resistant/tolerant varieties.

MATERIALS AND METHODS

Site and design

The experiments were conducted at net house and field of Plant Pathology Division, Bangladesh Rice Research Institute (BRRI), Gazipur, during the period from March to June 2008 for upland varieties and June to October 2008 for lowland rice varieties. In the field, the experiments were set up in randomized complete block design (RCBD) with three replications.

Plant materials

A total of 16 rice varieties including six uplands, eight lowlands, one resistant and one susceptible check were used in these studies (Table 1). The varieties were selected on the basis that they were the promising upland and rainfed lowland varieties for cultivation in the farmers field. The Kumragoir is a local rice variety and highly resistant, while Purbachi is a highly susceptible variety against tungro disease (Latif *et al.,* 2011). The seeds of all varieties were collected from Adaptive Research, and Genetic Resources and Seed Division, BRRI, Gazipur, Bangladesh.

Seedling growth

Sundried seeds of each variety were placed into petridish on one piece of filter paper. The filter paper was soaked with water to provide moisture for seed germination. The petridishes were kept in room temperature and water was added everyday to maintain moisture. Just after germination, seeds were sown in line in trays. One line was allocated for each variety and each line consisted of about 50-60 seeds. Each variety was sown in three trays. After ten days, two trays were used for inoculation and the third tray was used as control.

Inoculation of seedlings (at net house)

Initially GLHs were allowed to feed on 45-to 60-day-old infected source plants (affected by tungro disease) with distinct symptoms for 2 to 3 days. Ten-day-old seedlings of two trays of each variety were inoculated by viruliferous GLH for 24 hrs in wood-net made cage in net house. To ensure proper inoculation at least 3-4 viruliferous GLH per test plant were introduced into the cage. To ensure enough inoculum for each seedling the GLHs used for inoculation inside the case were disturbed several times during the inoculation access period. Three weeks after inoculation, symptoms appeared and the seedlings were scored based on visual

Table 1. Rice varieties of upland and rainfed lowland condition, 2008.

Variety	Eco-type	Reaction against tungro
	Upland	
BR8	Upland	Unknown
BR26	Upland	Unknown
BRRI dhan27	Upland	Unknown
BRRI dhan42	Upland	Unknown
BRRI dhan43	Upland	Unknown
BRRI dhan48	Upland	Unknown
	Lowland rainfed	
Purbachi	Upland/lowland	Susceptible check
Kumragoir	Upland/lowland	Resistant check
BRRI dhan33	Lowland	Unknown
BRRI dhan34	Lowland	Unknown
BRRI dhan37	Lowland	Unknown
BRRI dhan38	Lowland	Unknown
BRRI dhan39	Lowland	Unknown
BRRI dhan40	Lowland	Unknown
BRRI dhan41	Lowland	Unknown
BRRI dhan46	Lowland	Unknown

observation. Healthy checks were used to compare with diseased one.

Field experiment

The inoculated seedlings along with the control plants of each variety were transplanted in the field of Plant Pathology Division, BRRI. For each variety 1 m × 1 m plot was used. Row to row and plant to plant distance was maintained as 20 cm. The upland varieties were planted on high land with almost no supply of irrigation (limited irrigation was supplied only when needed) during March to June 2008. The rainfed varieties were planted when there was adequate rainfall to ensure water supply for plants during June to October 2008. Management practices such as weeding, fertilization were followed by BRRI recommendations (BRRI, 2000).

Data collection and statistical analysis

Data were collected on seedling infection (symptom), plant height (cm), number of tillers/plant, number of panicles/plant, panicle length (cm), number of filled grains/panicle and yield/plant (g) (Table 2). Seedling infection score was recorded from 10 inoculated plants by visual observation. From each replication five plants were selected randomly to record data for plant height, number of tillers/plant, number of panicles/plant, panicle length (cm), number of filled grains/panicle and yield/plant (g). The recovering ability was measured from the inoculated varieties by all the yield contributing parameters, implied how the infected plants showed their potentiality to provide better yield after being infected by tungro virus. The analysis of variance (ANOVA) which expressed the main interaction effect was analyzed by Statistical Analysis System (SAS version 9.2) for all quantitative traits. The Least Significant Difference (LSD) was performed for mean comparison when varietal differences were found significant at p=0.05 level of probability.

RESULTS

Upland rice

Recovering data. The morphological and yield traits varied significantly among the tested varieties in the present study (Table 3). The seedling infection by RTD was the highest in variety BRRI dhan27 (93.39%) followed by BRRI dhan43 (91.91%) and BR26 (91.59%) which were higher than the susceptible check Purbachi (90.39%) (Table 4). The resistant check Kumragoir did not show any symptoms of tungro disease in inoculated plants. The lowest seedling infection was found in variety BR8 (76.74%). All the eight varieties showed higher plant height (PH) in control plants than inoculated one. Among tested varieties the highest PH was recorded for the variety BR8 (45.76 cm) in inoculated plants, however in control plants this value was recorded as 92.13 cm, a reduction of 50.33% in this variety (Fig. 1). The lowest PH was observed in the variety BR26 (33.71 cm) in treated plants, while in control plants it was

Table 2. List of traits and their evaluation method.

Trait	Evaluation method
Seedling infection (%)	The yellow/orange yellow, twisted leaves were rated by visual observation
Plant height (cm)	Plant height was measured during harvesting from base of the plant to tip of the tallest leaf blade
Number of tillers/plant (no.)	Total number of tillers were counted from each plant where secondary and tertiary tillers were also included
Number of effective tillers/plant (no.)	Effective tillers were counted as those tillers which bear panicle.
Panicle length (cm)	The panicle length was recorded as the distance between the top most node of culm and the apex of panicle excluding awn.
Number of filled grains/panicle (no.)	Only the fertilized filled grains or fully developed spikelets were counted from each panicle of a plant and average of all panicles were recorded.
Yield/plant (g)	Total filled grains of a plant were weighed by an electric balance.

Table 3. Mean squares of analysis of variance for morphological and yield contributing characters studied in inoculated and uninoculated plants in upland rice varieties.

SV	df	SI	PH	NT/H	NP/H	PL	FG/P	Y/P
Block (B)	2	27.45 ^{ns}	2245.71*	3.66 ^{ns}	4.12 ^{ns}	3.48 ^{ns}	520.75 ^{ns}	0.80 ^{ns}
Variety (V)	7	1499.68**	1011.27 ^{ns}	68.36**	40.20**	37.65**	2862.67**	647.72**
Treatment (T)	1	71251.98**	23392.44**	0.73 ^{ns}	185.81**	92.66**	6143.14**	1055.25**
Var*Trt	7	1499.68**	83.02 ^{ns}	4.53 ^{ns}	5.62 ^{ns}	1.16 ^{ns}	1321.96**	60.49**
Error	30	44.66	414.14	4.66	5.56	1.37	365	0.68

SV, source of variation; df, degree of freedom; SI, % seedling infection; PH, plant height (cm); NT/H, total number of tillers plant^{-1}, NP/H, number of panicles/hill; PL, panicle length; FG/P, number of filled grains/panicle; Y/P, yield/plant(g) *Significant at 5% level; ** highly significant at 1% level; ^{ns} non significant.

Table 4. Morphological and yield contributing characters studied in inoculated and uninoculated plants in upland rice varieties (average of three replications).

Variety	% seedling infection		Plant height (cm)		No. of tillers/ hill		No. of panicles/ hill		Panicle length (cm)		No. of filled grains/panicle		Yield (g/plant)	
	Ino	Con	Ino	Con	Ino	Con	Ino	Con	Ino	Con	Ino	Con	Ino	Con
Purbachi	90.39	0.00	37.39	78.24	16.07	16.70	7.34	12.67	20.61	23.66	72.33	134.22	5.00	19.50
Kumragoir	0.00	0.00	138.33	143.89	16.63	15.93	11.67	15.56	27.56	28.93	117.78	144.00	50.00	62.00
BR8	76.74	0.00	45.76	92.13	7.77	10.37	4.33	7.78	18.63	21.79	76.00	52.22	10.82	18.33
BR26	91.59	0.00	33.71	74.50	12.67	10.77	3.55	11.67	19.29	22.91	68.56	91.22	3.42	9.22
BRRI dhan27	93.39	0.00	48.13	102.16	6.83	6.47	4.34	6.78	20.99	24.63	63.44	83.22	3.17	6.91
BRRI dhan 42	83.33	0.00	44.12	83.73	10.10	12.90	5.86	8.78	19.16	22.58	75.44	83.44	6.37	11.63
BRRI dhan 43	91.91	0.00	41.71	76.13	11.07	11.47	7.44	10.33	20.46	22.69	67.89	69.22	5.51	12.13
BRRI dhan 48	89.09	0.00	34.10	75.30	14.80	13.30	8.56	11.00	21.71	23.46	51.33	116.22	5.45	14.12
CV (%)	17.35		30.95		17.83		27.41		1.17		19.11		6.37	

Ino: inoculated plants (treated plants) and Con: control plants (uninoculated plants).

74.50 cm, a reduction of 54.75% compared to a 52.2% reduction in the susceptible check Purbachi. The lowest PH reduction was found in BRRI dhan43 (45.21%). In the resistant check Kumragoir PH reduction in inoculated plants was only 3.86% compared to control plants. In case of number of tillers/hill the variety BRRI dhan48 (14.80) produced more tillers among tested varieties in inoculated plants and BRRI dhan27 (6.83) produced less number of tillers/

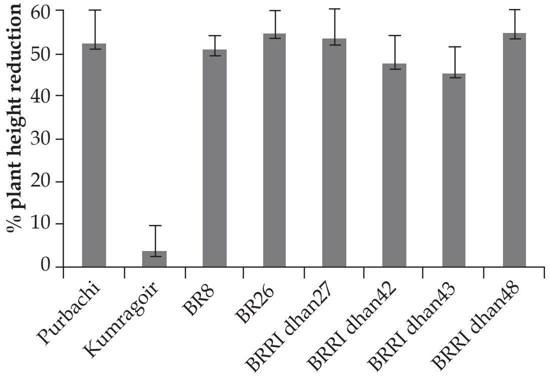

Fig. 1. Percent reduction of plant height in inoculated over control plants (upland).

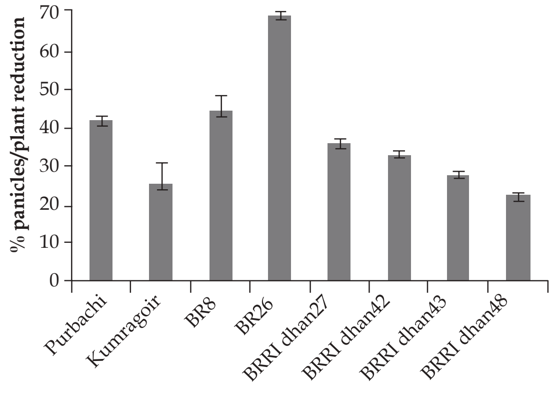

Fig. 2. Percent reduction of panicle numbers/hill in inoculated over control plants (upland).

hill. In control plants these two varieties had also the highest and lowest number of tillers/hill (13.30 and 6.47) respectively. Varieties BR26, BRRI dhan27 and BRRI dhan48 had more tillers/hill in inoculated plants than control plants (Table 3).

Yield data

The panicle numbers/hill was higher in control plants than inoculated plants for all the tested varieties. Reduction of panicle number was the lowest in BRRI dhan48 (22.18%) followed by BRRI dhan43 (27.98%) and BRRI dhan42 (33.26%) (Fig. 2). The highest percent of panicle panicle number reduction was observed in BR26 (69.58%) followed by BR8 (44.34%) and BRRI dhan27 (35.99%). The variety BRRI dhan48 (22.18%) showed lower panicle number reduction than the resistant check Kumragoir (25.00%) and the variety BR26 (69.58%) had higher basical number reduction than the susceptible check Purbachi (42.07%). The panicle length (PL), number of filled grains/panicle and yield (g/plant) were found to vary from 18.63 cm, 51.33 and 3.17 g/plant for the varieties BR8, BRRI dhan48, BRRI dhan27 to 21.71 cm, 76.00 and 10.82 g/plant for the varieties BRRI dhan48, BR8 and BR8 respectively in inoculated plants. On the other hand, in control plants these values ranged from 21.79 cm, 52.22 and 6.91 g/plant for the varieties BR8, BR8 and BRRI dhan27 to 24.63 cm, 116.22 and 18.33 g/plant for the varieties BRRI dhan27, BRRI dhan48 and BR8 respectively. Of tested varieties under upland conditions, the highest PL reduction was observed in BR26 (15.80%) followed by BRRI dhan42 (15.15%), BRRI dhan27 (14.78%) and BR8 (14.50%); and the lowest was in the variety BRRI dhan48 (7.46%) (Fig. 3). Number of filled grains/panicle (FG/P) was also higher in control plot than inoculated one for all the tested varieties except BR8. All the eight varieties showed higher yield in control plot than inoculated plants. Percent yield reduction was varied from 40.97 to 62.91 in the present study. The lowest yield reduction belonged to BR8 (40.97%) while it was the highest for the variety BR26 (62.91%) (Fig. 4). The other tested varieties BRRI dhan27, BRRI dhan42, BRRI dhan43 and BRRI dhan48 had the 54.12, 45.23, 54.58 and 61.40 % yield reduction respectively compared to control plants. The resistant check Kumragoir showed only 19.35% and the susceptible check Purbachi exhibited 74.35% yield reduction in inoculated plants.

Rainfed lowland rice

Recovering data. The effect of variety and treatment was highly significant in the present study (Table 5). Among the ten varieties, seedlings of nine varieties were infected 100% by tungro disease after artificial inoculation except resistant variety Kumragoir. No disease

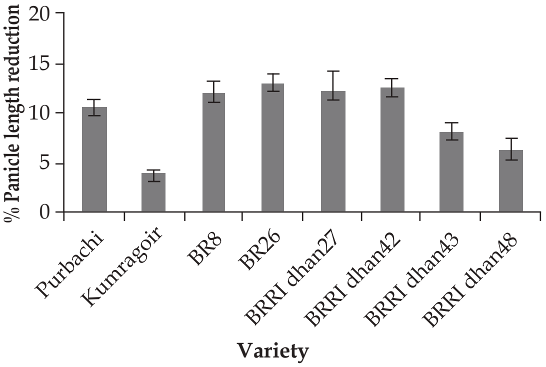

Fig. 3. Percent reduction of panicle length in inoculated over control plants (upland).

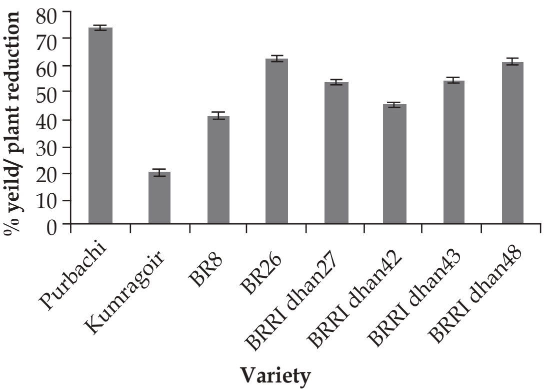

Fig. 4. Percent reduction of yield in inoculated over control plants (upland).

symptoms were observed in Kumragoir. All the ten varieties showed higher PH in control than inoculated plants (Table 6). The lowest PH reduction was observed in BRRI dhan46 (4.45%), which was almost similar to resistant check Kumragoir (3.86%) and the highest in the variety BRRI dhan33 (46.14%) (Fig. 5). The BRRI dhan37, BRRI dhan38 and BRRI dhan46 showed resistant reaction, while BRRI dhan33 and BRRI dhan40 revealed susceptible reaction against tungro disease in case of the character PH.

Yield data

Although the variety BRRI dhan46 had higher panicle length in inoculated (24.57 cm) plants than control (24.44 cm), the difference was not significant. On the contrary, number of filled grains/panicle and yield/plant was higher in control plants for this variety. Panicle length reduction was the lowest in BRRI dhan37 (0.32%) followed by BRRI dhan41 (3.21%) and BRRI dhan39 (12.53%) (Fig. 6). For the grain character number of filled grains/panicle was higher in healthy plants than diseased one for all the tested varieties. Reduction in filled grains/panicle of up to 100% was observed in

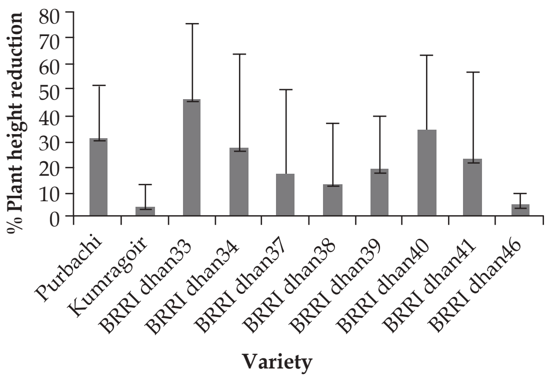

Fig. 5. Percent reduction of plant height in inoculated over control plants (rainfed lowland).

Table 5. Mean squares of analysis of variance for morphological and yield contributing characters studied in inoculated and uninoculated plants in lowland rice varieties.

SV	df	SI	PH	NT/H	NP/H	PL	FG/P	Y/P
Block (B)	2	0.00	14049.00**	1.78^{ns}	32.08*	0.08 ^{ns}	802.91 ^{ns}	0.71 ^{ns}
Variety (V)	7	1500.00**	3794.15**	99.64**	64.19**	103.57**	12310.51**	1084.20**
Treatment (T)	1	121500.00**	3597.71**	90.53**	159.54**	335.12**	56765.12**	291.06**
Var*Trt	7	1500.00**	101.74^{ns}	21.10*	5.99 ^{ns}	80.20**	3672.22**	196.89**
Error	30	0.00**	185.90	8.23	7.59	1.75	439.90	1.82

SV, source of variation; df, degree of freedom; SI, % seedling infection; PH, plant height (cm); NT/H, total number of tillers plant^{-1}, NP/H, number of panicles/hill; PL, panicle length; FG/P, number of filled grains/panicle; Y/P, yield/plan t(g) *Significant at 5% level; **highly significant at 1% level; non significant.

Table 6. Morphological and yield contributing characters studied in inoculated and uninoculated plants in lowland rainfed rice varieties (average of three replications).

Variety	% seedling infection		Plant height (cm)		No. of tillers/hill		No. of panicles/ hill		Panicle length (cm)		No. of filled grains/panicle		Yield (g/plant)	
	Ino	Con	Ino	Con	Ino	Con	Ino	Con	Ino	Con	Ino	Con	Ino	Con
Purbachi	100.00	0.00	43.35	62.09	6.33	17.10	6.33	8.56	19.21	21.00	42.44	87.67	8.89	12.00
Kumragoir	0.00	0.00	135.30	140.50	15.03	14.53	10.67	14.60	14.40	26.40	113.50	141.45	45.00	53.50
BRRI dhan33	100.00	0.00	32.15	59.69	2.23	10.27	0.44	5.56	13.52	19.24	6.67	54.33	0.13	5.86
BRRI dhan 34	100.00	0.00	59.22	80.93	13.70	14.57	5.56	5.78	22.48	25.73	121.56	239.56	3.78	10.60
BRRI dhan 37	100.00	0.00	67.22	80.93	15.97	17.23	7.65	8.78	22.07	22.14	139.00	185.44	15.20	17.77
BRRI dhan 38	100.00	0.00	68.07	78.18	20.53	21.90	10.67	12.22	20.76	25.84	71.89	120.67	15.55	23.00
BRRI dhan 39	100.00	0.00	52.61	64.36	12.67	13.47	0.89	6.11	20.32	23.23	60.89	74.78	5.83	8.20
BRRI dhan 40	100.00	0.00	50.43	76.19	12.57	12.97	3.11	5.89	0.00	24.79	0.00	177.44	0.00	24.20
BRRI dhan 41	100.00	0.00	60.42	77.78	11.97	12.67	2.33	6.45	24.70	25.52	84.78	147.11	4.44	26.40
BRRI dhan 46	100.00	0.00	65.24	68.28	18.0	18.87	2.78	9.11	24.57	24.44	88.67	116.11	15.83	28.20
CV (%)	0.00		13.63		20.31		41.27		6.17		20.22		7.18	

Ino: inoculated plants (treated plants) and Con: control plants (uninoculated plants).

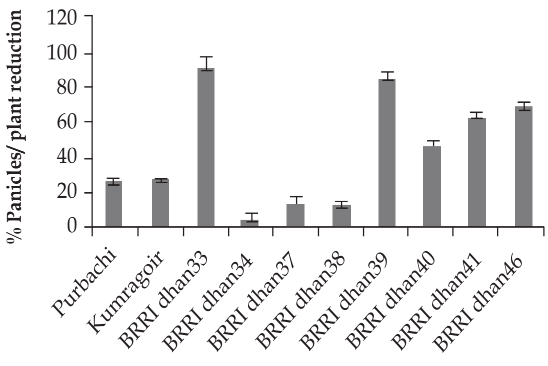

Fig. 6. Percent reduction of panicle numbers/hill in inoculated over control plants (rainfed lowland).

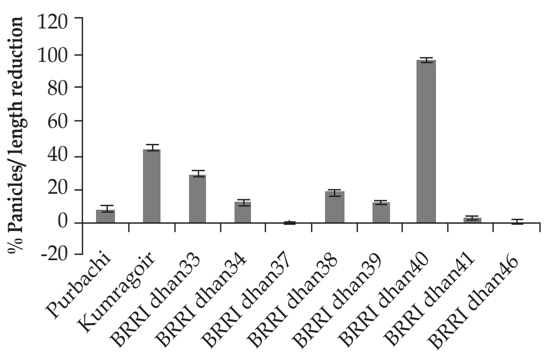

Fig. 7. Percent reduction of panicle length in inoculated over control plants (rainfed lowland).

the variety BRRI dhan40 (Fig. 7). All the ten varieties showed higher yield in control plot than inoculated one. In lieu of yield, BRRI dhan37 (14.46%) showed the lowest yield reduction. It showed even less yield reduction than the resistant check Kumragoir (15.89%), simultaneously exhibited the highest recovering ability in lowland condition (Fig. 8). The BRRI dhan33 and BRRI dhan40 had 97.78% and 100% yield reduction revealed these varieties did not have any recovering ability under tungro disease infection.

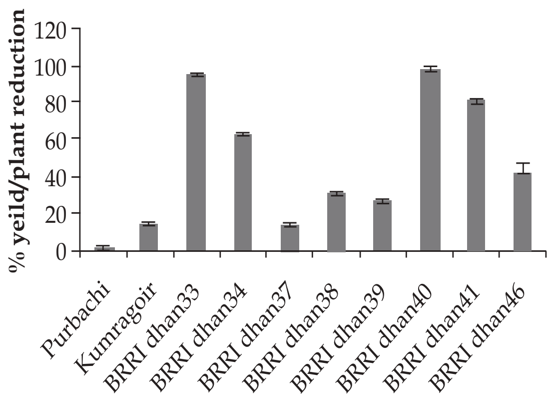

Fig. 8. Percent reduction of yield in inoculated over control plants (rainfed lowland).

DISCUSSION

Yellow or orange-yellow discoloration of leaves, stunting of plant growth, reduction in number of effective tillers, shortness of panicle length, and often sterile or partial filled grains are the characteristics of tungro disease in rice. In our experiment, we observed these kinds of symptoms in all tested inoculated varieties. Some varieties exhibited severe, while others showed mild tungro symptoms after inoculation. However, the varieties which had recovering ability were recovered up to harvest stage while some varieties had very less ability to recover against tungro disease.

Upland rice

In upland rice, all varieties showed better results in healthy plants than diseased plants for the agronomic character PH. It is supported by Agrios, 2005. He stated tungro-infected rice plants are stunted compared to healthy rice plants. Some varieties, such as Kumragoir, BR26, BRRI dhan27 and BRRI dhan48 showed higher number of tillers/hill in diseased plants. In contrast, productive tillers were always higher in control plants than diseased treatment for all varieties. One of the characteristics of tungro disease is that the infected plants become bushy than uninfected plants. In this study, we observed this symptom in treated plants. Although diseased plants had more tillers but most of them were secondary and tertiary and so they did not bear any panicle. Panicle length was also higher in healthy plants for all varieties than diseased plants. The panicles of infected plants are often small, sterile and incompletely exerted (www.rkmp.co.in/cont/symptoms-of-rice-tungro-disease). All varieties showed higher number of filled grains/panicle in control plants than inoculated plants except BR8, which had more filled grains in diseased plants than healthy ones and consequently it showed lowest yield reduction and had highest recovering ability in terms of yield (g/plant). The BRRI dhan42 and BRRI dhan27 showed moderately tolerant reaction ie moderate recovering ability against tungro disease. During the screening of BRRI rice varieties Latif *et al*. (2011) also found the similar result for BRRI dhan27 as in the present study. Among tested varieties, BR26 exhibited the lowest recovering ability for the character yield/plant (g). The same result was also reported in a study conducted by BRRI (2000) that BR26 is susceptible to rice tungro disease.

Rainfed lowland rice

The BRRI dhan33 and BRRI dhan40 did not show any tolerance or recovering ability. BRRI dhan34 and BRRI dhan41 had some extent tolerance, which showed slight recovery against tungro. BRRI dhan37 and BRRI dhan39 exhibited more tolerance or higher recovering ability for both number of filled grains/panicle and yield (g/plant), although all these varieties seedlings were 100% tungro infected. In a similar study, Latif *et al*. (2011) found that BRRI dhan33 is moderately susceptible while BRRI dhan37 are moderately resistant against rice tungro disease. Among susceptible varieties, BRRI dhan40 did not produce any yield; although it had some panicles, all spikelets were sterile, which is one of the most prominent symptoms of tungro disease. In contrast, BRRI dhan37 showed strong recovering ability and the highest tolerance after 100% seedlings being affected by tungro, which showed less reduction of number of panicles/hill, panicle length and yield (g/plant), even than the resistant variety Kumragoir. Therefore, this variety could be used as a parental line in crossing programme for the development of tungro resistant varieties or could be used in tungro endemic areas where it would provide better yield although affected by tungro disease.

Resistant to tungro disease has been an important breeding objective for rice improvement in many Asian countries (Ling, 1974; Anjanejulu *et al.,* 1982; Buddenhagen, 1983). Many cultivars bred as tungro-resistant had resistance to GLH (Rapusas and Heinrichs, 1982; Heinrichs and Rapusas, 1983; Hibino *et al.,* 1987) and did not last long (Inoue and Ruy-Aree, 1977; Manwan *et al.,* 1985; Hibino *et al.,* 1987; Dahal *et al.,* 1990). High yielding cultivars with resistance to tungro have succumbed after a few years of intensive cultivation in Indonesia (Manwan *et al.*, 1985;), Philippines (Hibino *et al.*, 1987; Dahal *et al.*, 1990) and Thailand (Inoue and Ruy-Aree, 1977). For this reason, it is pertinent to identify varieties with higher tolerance or high recovering ability. In the present case, all the varieties were artificially inoculated by viruliferous GLH. All varieties showed distinct tungro symptom after three weeks of inoculation and also after transplanting in the field. With the advancement of plant growth, varieties BR8 and BRRI dhan37 recovered from tungro syndrome due to their genetic makeup consisting tolerance potentiality showed better yield, while other varieties like BRRI dhan33 and BRRI dhan40 did not return any yield.

CONCLUSION

Among upland, BR8 and among lowland, BRRI dhan37 were identified as tolerant varieties, which exhibited highest recovering ability in respect to character number of panicles/hill, panicle length and yield (g/plant), could be cultivated in tungro disease endemic areas. In these studies, all test varieties were artificially inoculated by viruliferous GLH. All the varieties showed typical tungro symptom after 21 days of inoculation and also in the field. But with the progress of plant development, varieties BR8 and BRRI dhan37 recovered from tungro syndrome due to their genetic makeup consisting tolerance potentiality showed better yield, at the same time other varieties, for example, BRRI dhan33 and BRRI dhan40 had no yield return.

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Short Communication

Effect of Leaf Clipping on Yield Attributes of Modern and Local Rice Varieties

S R Das[1], M Y Ali[1,*] and M M Islam[1]

ABSTRACT

Impact of five different types of leaf clipping on the yield attributes of modern (Binadhan-8) and local (Terebaile) rice variety was evaluated on pot experiments following a completely randomized design (CRD) with three replications. Leaves were cut according to the treatment. Data were collected on panicle length (cm), filled grain panicle^{-1}, unfilled grain panicle^{-1}, thousand grain weight (g), grain weight panicle^{-1} (g). In Binadhan-8, flag leaf alone or flag leaf with 2nd leaf and 2nd and 3rd leaves cutting showed profound reduction in grain number panicle^{-1} (35.14, 62.62, and 51.83%, respectively) and grain weight panicle^{-1} (29.18, 58.37 and 48.93%, respectively) while, cutting of 2nd leaf and 3rd leaf alone exert no significant impact compared to control. Number of unfilled grain increased with higher intensity of leaf cutting. In Terebaile, only flag leaf cut showed non-significant impact on grain number panicle^{-1} and grain weight panicle^{-1}. Profound impact was observed by cutting flag leaf with 2nd leaf (55.47 and 48.98%, respectively) and flag leaf with 2nd and 3rd leaf (58.96 and 63.13%, respectively). Leaf clipping had non-significant effect on thousand grain weight of modern variety Binadhan-8 while, it had significant effect in Terebaile.

Key words: Leaf clipping, rice variety, yield attributes

INTRODUCTION

Rice (*Oryza sativa* L.) is the staple food of about 166 million people of Bangladesh. Rice contributes about 60% GDP of the crop sector in Bangladesh and employs 47.8% of the total labour force (www. worldometers.info).

In cereal crops, yield formation is a complex process. Leaves are vitally essential organs for photosynthesis, which is a major process affecting crop growth rates and is affected by either the number or the area of the leaves. Since the productivity of a plant depends on the efficiency of its photosynthetic processes, the growth and development of leaves (Karadogan and Akgun, 2009). Leaf has important role to improve yield. During grain formation stage, the youngest flag leaf of rice remained metabolically more active. The grain yield and straw yield of rice increased significantly with the presence of flag leaf.

Leaf photosynthesis can be influenced by many plant factors such as leaf age, leaf position, sink effects, and mutual shading, as well as environmental factors such as light, temperature, nutrition, and water availability (Lieth and Pasian, 1990). Defoliation or leaf damage can decreases assimilate availability during grain filling, seed and biological yield (Echarte *et al*., 2006).

Tiller number and flag leaf play an important role regarding growth and yield of rice. About 60-90% of the total carbon content in the panicles at harvest is obtained from photosynthesis after heading while around equal or more than 80% of nitrogen in the panicles at harvest is absorbed before heading and remobilized from vegetative organs (Mae, 1997). Misra (1986) observed both the

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economic and biological yields were closely related with optimum leaf area index of plant population of different rice varieties. Flag leaf contributed to 45% of grain yield and is the single most component for yield loss (Abou-khalifa *et al*., 2008).

The top three leaves not only assimilate majority of carbon for grain filling during ripening phase but also provide large proportion of remobilized-nitrogen for grain development during their senescence (Misra and Misra, 1991; Mae, 1997).

The grain yield and straw yield of rice increased significantly with the presence of flag leaf. Local rice variety differs from modern variety in various morphological and physiological aspects specifically in transformation of assimilation to grain development.

Considering the facts discussed above the present research work has been undertaken to determine the effect of leaf clipping on yield attributes of modern rice variety Binadhan-8 and local rice variety Terebaile.

The experiment was conducted in the net house of Plant Breeding and Biotechnology Laboratory of Khulna University, Khulna during the Boro season 2013 (January–June). Rice cultivars Binadhan-8 (modern) and Terebaile (local) were. Following leaf clipping treatments were applied for both the experiments.

L_0- Control (without leaf cutting)

L_1 - Flag leaf cut

L_2- 2nd leaf cut

L_3- 3rd leaf cut

L_4- Both flag leaf and 2nd leaf cut

L_5- Flag leaf with 2nd and 3rd leaves cut together

The experiments were laid out in a completely randomized design (CRD) with three replications.

Earthen pots of 11″(27.94 cm) in upper diameter and 9″(22.86 cm) in height were used for the experiments. Every pot was filled with 8 kg of dry soil treated with bleaching powder @ 100 mg per kg soil to kill soil borne pathogen. Fertilizer doses were 0.22, 0.13, 0.08, 0.03 and 0.14g urea, TSP, MoP, zinc sulphate and Gypsum respectively for kg^{-1} dry soil.

One-third of urea and full doses of other fertilizers were applied as basal doses and then incorporated to soil by hand. Rest of the two-third urea was applied in two equal splits at 15 and 45 days after transplanting. Forty-three-day-old seedlings were transplanted at the rate of single seedling in each pot on 7 February 2013. Irrigation and other intercultural operations were done as and when required. Leaf clipping was done prior to panicle emergence stage following the assigned treatment. Binadhan-8 and Terebaile were harvested on 28 April 2013 and 10 May 2013 respectively. Yield contributing data were recorded as follows: panicle length, grain number panicle^{-1}, unfilled grain number panicle^{-1}, thousand grain weight (TGW), grain weight panicle $^{-1}$. All the data were analyzed by MSTAT-C. The differences between the treatment means were determined by using Duncan's new multiple range test (DMRT).

Significant effect of leaf clipping was noticed in most of the parameters studied in both the rice cultivars. In Binadhan-8, the highest number of filled grain panicle^{-1} was found in control plants (104.00) which did not vary significantly from 2nd and 3rd leaf cut. Significant reduction in filled grains takes place by flag leaf cut (35.14%), flag leaf with 2nd leaf cut (62.62%) and flag leaf with 2nd and 3rd leaf cut (51.83 %). Unfilled grain number increased with higher intensity of leaf cutting and was the highest (79.40) in flag leaf with 3rd leaf cut (133.59%), which was similar with flag leaf with 2nd leaf cut (65.91). The lowest unfilled grain was in the control (33.99) which did not vary with 3rd leaf cut alone (39.57). Flag leaf cut and 2nd leaf cut showed the similar and moderate values (Table 1). TGW did not vary due to leaf cutting. Grain weight panicle^{-1} was the highest in control plants (2.33 g), which was not affected due to 2nd leaf and 3rd leaf cut. Grain weight significantly reduced due to flag leaf cut (29.18%), flag leaf with 2nd leaf cut (58.37%) and flag leaf with 2nd and 3rd leaves cut (48.93%).

Table 1. Effect of leaf clipping on yield attributes of modern rice cultivar Binadhan-8.

Treatment (Leaf clipping)	Panicle length (cm)	Filled grain panicle $^{-1}$	Thousand grain weight (g)	Grain weight panicle $^{-1}$ (g)
L_0	24.97	104.00 a	21.37	2.33 a
L_1	20.65	67.45bc (35.14)	21.57	1.65bc (29.18)
L_2	24.93	95.37ab (8.29)	20.04	2.11ab (9.44)
L_3	26.11	91.60ab (11.92)	21.73	2.12ab (9.01)
L_4	23.27	38.88c (62.62)	20.44	0.97d (58.37)
L_5	23.56	50.10c (51.83)	20.66	1.19cd (48.93)
CV (%)	7.85	15.57	6.61	14.22
Level of significance	NS	0.01	NS	0.01

Data in a column accompanied by the same letter (s) are not significantly different at the 1% level as per DMRT. Figures in the parenthesis indicate the percent reduced / increased over the control. CV= Coefficient of variation; NS= Not significant. L_0- Control (without leaf cutting), L_1 - Flag leaf cut, L_2- 2nd leaf cut, L_3- 3rd leaf cut, L_4- Both flag leaf and 2nd leaf cut, L_5 - Flag leaf with 2nd and 3rd leaves cut together.

In Terebaile, leaf clipping exhibits significant effect on yield attributes except panicle length. Number grain panicle^{-1} did not vary from control due to flag leaf cut and 3rd leaf cut. Number of filled grain reduced significantly due to 2nd leaf cut (18.57%), flag leaf with 2nd leaf cut (55.47%) and flag leaf with 2nd and 3rd leaves cut (58.96%). Number of unfilled grain increased with higher intensity of leaf cutting. The highest number of unfilled grain was in flag leaf with 2nd leaf cut (76.94) which was 482.44% higher than control (13.21). Rest of the treatments had statistically similar values with the control (Table 2). TGW decreased with the progressive increase in leaf clipping and the highest value was in the control treatment (22.20 g) and the lowest value (15.27 g) was in flag leaf with 2nd and 3rd leaves cut (31.22% less). Only flag leaf, 2nd leaf and 3rd leaf cut exhibits no significant impact on TGW compared to control rather than flag leaf together with 2nd leaf as well as 2nd and 3rd leaves cut. Grain weight

Table 2. Effect of leaf clipping on yield attributes of local rice cultivar Terebaile.

Treatment (Leaf clipping)	Panicle length (cm)	Filled grain panicle $^{-1}$	Thousand grain weight (g)	Grain weight panicle $^{-1}$ (g)
L_0	21.24	92.55ab	22.20a	1.98a
L_1	20.84	86.46ab (6.58)	21.23ab (3.92)	1.95a (1.52)
L_2	19.63	75.36b (18.57)	19.95ab (10.14)	1.59ab (19.69)
L_3	22.62	100.7a (8.81)	20.27ab (8.69)	2.10a (6.06)
L_4	21.25	41.21c (55.47)	17.92bc (19.28)	1.01bc (48.98)
L_5	20.67	37.98c (58.96)	15.27c (31.22)	0.73c (63.13)
CV (%)	8.63	12.91	10.02	15.92
Level of significance	NS	0.01	0.05	0.01

Data in a column accompanied by the same letter (s) are not significantly different at the 1% level as per DMRT. Figures in the parenthesis indicate the percent reduced / increased over the control. CV= Coefficient of variation; NS= Not significant. L_0- Control (without leaf cutting), L_1 - Flag leaf cut, L_2- 2nd leaf cut, L_3- 3rd leaf cut, L_4- Both flag leaf and 2nd leaf cut, L_5 - Flag leaf with 2nd and 3rd leaves cut together.

panicle^{-1} was not changed due to flag leaf, 2nd leaf and 3rd leaf cut alone compared to control (1.98 g). Flag with 2nd leaf and flag leaf with 2nd and 3rd leaf cut showed marked reduction in grain weight panicle^{-1} where the latter one bears the lowest value (0.73 g), which was 63.13% less than the control.

DISCUSSION

Leaf clipping effects significantly on the yield attributes of both modern and local rice varieties. In the modern variety, the impact of flag leaf cut was prominent rather than 2nd and 3rd leaf cut alone but profound impact was observed by flag leaf together with 2nd leaf and 2nd as well as 3rd leaves cut. It indicates that, flag leaf plays a vital role in the supply of assimilate to panicle in absence of 2nd leaf or 2nd as well as 3rd leaves. This result is in agreement with earlier reports that, flag leaf contributed to 45% of grain yield (Misra, 1995) and is the single most component for yield loss (Abou-Khalifa *et al.,* 2008). In local variety, number of filled grain and grain number panicle^{-1} did not vary significantly in absence of flag leaf compared to the control. It indicates that 2nd and 3rd leaves can compensate for the flag leaf. Marked impact was noticed by cutting flag leaf together with 2nd leaf and 2nd as well as 3rd leaves. Similar finding was also reported by Misra and Misra (1991) and Mae (1997), who stated that the top three leaves not only assimilate majority of carbon for grain filling during ripening phase but also provide large proportion of remobilized-nitrogen for grain development during their senescence. Abou-Khalifa *et al.* (2008) showed that in rice leaf clipping, L5, L4, L1, L2 and L3 treatments the loss of grain yield was 59.87, 94.92, 44.89, 29.58 and 19.98% of control, respectively. Leaf cutting exhibits non-significant impact on TGW of modern variety but it was significant in local variety. This result was supported by Abou-Khalifa *et al.* (2008) who stated that in case of leaf cutting, hybrid rice cultivar H5 had relatively higher but non-significant TGW than the traditional inbred Egyptian local rice cultivar Sakha 103.

CONCLUSIONS

In modern variety, flag leaf plays a vital role in grain development, which could not be substituted by 2nd and 3rd leaves but in local variety, 2nd and 3rd leaves can be an effective substitute for grain development in absence of flag leaf. Leaf clipping had non-significant effect on TGW of modern variety while it was significant in local variety.

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