Complete submergence up to 1-2 weeks of low land rice crops during flash floods occurs in large areas of South-East Asia including Bangladesh, resulting in increased mortality of plants as well as low productivity of rice crops. A pot experiment was conducted at the Net House, Department of Crop Botany, Bangladesh Agricultural University, Mymensingh, during Aman season from July 2017 to December 2017 to evaluate morphological and phenological traits associated with submergence tolerance in rice. The experiment consisted of two factors—Rice cultivars (Binadhan-11, Binadhan-12, BRRI dhan51 and BRRI dhan52 as tolerant and BRRI dhan49 as susceptible) and submergence stress for 14 days at vegetative stage and control. Submergence stress was imposed by dipping of pots into a water tank with about 90 cm depth of water. After desubmergence, the plants were grown with proper care till maturity. Control plants are maintained in the pot house of field laboratory. Leaf greenness was measured after desubmergence to physiological maturity. The tolerant cultivars maintain higher leaf greenness for a long time than the susceptible cultivar after desubmergence. Reduction of grain filling rate and yield was significantly higher in susceptible cultivar than the tolerant cultivars. Among the five cultivars, BRRI dhan51 contributed the highest yield under stress treatment. Submergence tolerant rice cultivars (Binadhan-11, Binadhan-12, BRRI Dhan51 and BRRI Dhan52) had maintained higher tiller number, 1000-grain weight and the higher number of grains per panicle during submergence, as compared to susceptible rice cultivar BRRI Dhan49.

Introduction

Rice (Oryza sativa L.) belonging to the family Poaceae and subfamily Oryzoideae is the staple food of the people of Bangladesh. It is the second most important cereal crop in the world (FAO, 2017). World rice production in 2017 was 759.6 million tonnes (503.9 million tonnes, milled basis) with an increase of 0.6% from previous year (FAO, 2017). Rice production in Bangladesh is estimated at 36.27 million tons in the area under 11.62 million hectares in 2018 (BBS, 2018). Rice production has been badly affected in Bangladesh as well as throughout the world due to flooding. So it is a major thirst to develop submergence tolerant rice cultivars to meet the present food demand throughout the world. A submerged plant is defined as “a plant standing in water with at least part of the terminal above the water or completely covered with water” (Nishiuchi et al., 2012). Submergence subjects plants to the stresses of low light, limited gas diffusion, effusion of soil nutrients, mechanical damage, and increased susceptibility to pests and diseases (Ram et al., 2002). In addition to the O₂ deficiency, production of toxic substances such as Fe³⁺, Mn⁴⁺, and H₂S by reduction of redox potential causes severe damage to plants under waterlogged conditions (Drew and Lynch, 1980).

Unlike other crop plants, rice has some adaptive traits for tolerance of submergence. One of the traits is the formation of the longitudinal interconnection of gas spaces called aerenchyma that enables internal aeration between shoot and roots (Armstrong, 1980; Colmer, 2003; Colmer and Pedersen, 2008a). Moreover, leaf gas films, which are a micro-layer of air trapped between submerged leaves and the surrounding water, contribute to the internal aeration during submergence, thereby increasing submergence tolerance in rice (Colmer and Pedersen, 2008b).

So far, a limited number of high yielding varieties tolerant of submergence have been developed in Bangladesh to mitigate the crop loss due to submergence. However, the detailed morphological and phenological traits of the cultivars associated with the submergence are still poorly understood. Therefore, this study was carried out to examine the effects of submergence stress on grain yield, leaf greenness along with phenological traits of different rice cultivars.
Materials and Methods

Experimental Materials

Five cultivars of rice of which one submergence susceptible BRRI dhan49 and other four submergence tolerant rice cultivars BRRI dhan51, BRRI dhan52, Binadhan-11 and Binadhan-12 were used for the study. Seeds of BRRI dhan49, BRRI dhan51 and BRRI dhan52 were collected from Plant Breeding Division of Bangladesh Rice Research Institute (BRRI). Seeds of Binadhan-11 and Binadhan-12 were collected from Bangladesh Institute of Nuclear Agriculture (BINA).

Conduction of the experiment

The experiment was conducted during the period from July 2017 to December 2017 in plastic pots at the Net House, Department of Crop Botany, Bangladesh Agricultural University, Mymsenhing. The study was laid out as a two-factor experiment in completely randomized design (CRD) with 10 treatments and 3 replications. Factor A: Five rice cultivars (Binadhan-11, Binadhan-12, BRRI dhan51 and BRRI dhan52 as tolerant and BRRI dhan49 as susceptible) and Factor B: Submergence stress (control and submerged condition).

A total of 100 pots were prepared as it was a destructive method both for submergence stress and control. For sampling, we made out the plants from the submergence stress and after recording data we did not place it again to the water tank. Well-pulverized soil was mixed with well-decomposed cow dung properly. The plastic pots were filled with soil and cow dung mixture (2:1). Each pot contained about 10 kg of soil and cow dung mixture. Seeds of five cultivars were sown at five different trays on 20 June, 2017. The 20 days old seedlings were transplanted into the prepared pots on 10 July, 2017. Fertilizers were applied in the experimental pots as recommended dose. Weed free condition was maintained throughout the season. Watering was done when needed. Insecticide and pesticide were applied to maintain insect and disease free condition.

The water tank was partially filled with tap water of about 0.5 m height and about 45 pots with rice plants (9 pots of each rice cultivar) were dipped in the water of the tank one by one gently. The height of the tank water was raised gradually to submerge the plants completely. The light intensity of the tank water at 15 cm and 30 cm depth were measured 65.72 and 47.32 µmol by Light meter (Model- LI- 250A, Ligor Bio-Sciences, USA). The light intensity of air also measured and it was 240 µmol. Dissolved oxygen concentration was measured by DO meter (HANNA Instrument, Germany) and it was 10.50 ppm by DO meter. The temperature of the water was measured as 30.8 °C. The water height was kept constant by the daily supply of tap water. After 14 days of submergence, the pots with plants were removed from the water tank after draining the water from the tank using a siphoning pipe. Then the pots were placed in a sunny and restricted place and were allowed the plants to be established successfully.

Sampling

Grain filling period was defined as the duration from anthesis to physiological maturity. The date of anthesis was determined when the anthers extruded in 50% of the panicles in the field and the date of physiological maturity was the day when the grain attained its maximum weight. The days to anthesis was calculated as the duration from sowing to anthesis. The grains were separated from the panicles with tweezers and weighed. The average rate of grain filling was estimated as maximum grain weight divided by the duration (Julian days), assuming that grain weight was zero at anthesis (Dias and Lidon, 2009).

Estimation of leaf greenness

The greenness of the flag leaves (or topmost leaves in absence of flag leaves) was measured at different days after desubmergence in all cultivars with a chlorophyll meter (SPAD-502; Konica Minolta Sensing Inc., Osaka, Japan). SPAD readings were taken at three positions (near base, near apex and middle) of the leaf and were averaged for each leaf. Three leaves were measured and the readings were averaged for a pot.

Harvesting and data recording

The plants were harvested at maturity for yield and yield components at different dates for control and different days after submergence for stressed plants. The panicles were counted and the plants were dried for 48 h at 80°C and weighed to determine the biomass yield. All panicles were hand-threshed and weighed for grain yield. One thousand grains were counted and weighed to determine 1000-grain weight. The harvest index was calculated as the grain yield divided by total dry mass.

Statistical analysis

All data on yield and yield components were subjected to two factors (cultivar, submergence stress) analysis of variance in completely randomized design (CRD). All data were analyzed statistically using MS Office Excel application.

Results

Yield and yield components

The grain yield, biomass yield, harvest index and yield components of five rice cultivars as affected by submergence stress at the early vegetative stage were shown in Table 1. The submergence stress had a significant effect on grain yield. The grain yield varied from 16.09 to 23.36 g pot⁻¹ with the mean of 20.90 g pot⁻¹ under control while it varied from 0.82 to 21.32 g.
pot\(^{-1}\) with the mean of 14.60 g pot\(^{-1}\) under submergence stress. The order of the cultivars in respect to grain yield under control condition was as follows: BRRI Dhan49 (23.36 g pot\(^{-1}\)) > BRRI Dhan51 (23.06 g pot\(^{-1}\)) > BRRI Dhan52 (21.17 g pot\(^{-1}\)) > Binadhan-11 (21.06 g pot\(^{-1}\)) > Binadhan-12 (16.09 g pot\(^{-1}\)). However, the order for grain yield under submergence stress was as follows: BRRI dhan51 (21.32 g pot\(^{-1}\)) > BRRI dhan52 (18.10 g pot\(^{-1}\)) > Binadhan-11 (17.39 g pot\(^{-1}\)) > Binadhan-12 (15.38 g pot\(^{-1}\)) > BRRI Dhan49 (0.82 g pot\(^{-1}\)). The biomass yield showed significant differences among the cultivars and the stress treatments. It ranged from 38.11 to 44.90 and 14.04 to 40.55 g pot\(^{-1}\) under control and stress treatment, respectively.

The number of panicles per pot exhibited significant differences among the cultivars but exhibited non-significant differences between the stress and control with the range of 6.33–9.00 and 1.00–10.00 under control and the submergence stress, respectively. The number of grains per panicle also exhibited significant differences among the cultivars and also between the stress and control treatments with the range of 93–208 and 81–145 under control and the stress, respectively.

The thousand grain weight varied significantly with the cultivars and also with the control and stress treatments. It varied from 15.96 to 27.20 g with the mean of 20.77 g and from 10.13 to 25.09 g with the mean of 17.60 g under control and stressed condition, respectively. The order of the cultivars according to grain weight under control was found as follows: Binadhan-11 (27.21 g) > BRRI Dhan52 (24.17 g) > BRRI Dhan49 (18.37 g) > BRRI Dhan51 (18.13 g) > Binadhan-12 (15.96 g). The order of the cultivars according to grain weight under the submergence stress as follows: Binadhan-11 (25.09 g) > BRRI Dhan52 (22.86 g) > BRRI Dhan51 (15.79 g) > Binadhan-12 (14.11 g) > BRRI Dhan49 (10.13 g).

### Phenological characters and grain filling rate

The phonological characters like days to anthesis, days to physiological maturity and grain filling duration; and average grain-filling rate of five rice cultivars under control and submergence stress were shown in Table 2. The grain-filling period (GFP) varied significantly with the cultivars but non-significantly with the control and stress treatments. It ranged from 34 to 45 with the mean of 39.4 under control. In submerged condition, GFP ranged from 37 to 44 with the mean of 39. BRRI Dhan49 possessed longest grain filling period (45 d) and Binadhan-11 possessed shortest period (34 d) under control while BRRI Dhan51 possessed longest grain filling period (44 d) and BRRI Dhan49, Binadhan-12 possessed shortest period (37 d) under submergence stress.

The days to anthesis showed significant differences among the cultivars and also between the control and submergence stress. It ranged from 93 to 111 and from 106 to 124 days under control and stress condition, respectively. The days to physiological maturity (PM) varied significantly with the cultivars and the stress treatments. It ranged from 127 to 150 and 145 to 168 days under control and stress condition. BRRI Dhan51 required the longest time (150 d) and Binadhan-11 required shortest time (127 d) to attain PM under control condition. In submerged condition BRRI Dhan51 required the longest time (168 d) and, Binadhan-11 and BRRI Dhan52 required the shortest time (145 d) to attain PM.

Table 1. Grain yield, biomass yield, and yield attributes in five rice cultivars as affected by submergence stress at early vegetative stage

| Cultivars     | Submergence stress | Grain yield/pot (g) | Biomass yield/pot (g) | No. of panicles/pot | No. of grains/pot | 1000-grain weight (g) |
|---------------|--------------------|---------------------|-----------------------|---------------------|------------------|----------------------|
| Binadhan-11   | Control            | 21.06               | 42.67                 | 9.00                | 93               | 27.21                |
|               | Stress             | 17.39               | 33.63                 | 9.00                | 81               | 25.09                |
| Binadhan-12   | Control            | 16.09               | 38.11                 | 7.00                | 165              | 15.96                |
|               | Stress             | 15.38               | 31.94                 | 10.00               | 128              | 14.11                |
| BRRI dhan51   | Control            | 23.06               | 44.90                 | 8.33                | 208              | 18.13                |
|               | Stress             | 21.32               | 40.55                 | 9.66                | 142              | 15.79                |
| BRRI Dhan52   | Control            | 21.17               | 40.39                 | 6.33                | 163              | 24.17                |
|               | Stress             | 18.10               | 34.14                 | 6.33                | 145              | 22.86                |
| BRRI Dhan49   | Control            | 23.36               | 41.50                 | 8.00                | 173              | 18.37                |
|               | Stress             | 0.82                | 14.04                 | 1.00                | 98               | 10.13                |

| Significance | Cultivar | ** | ** | ** | ** | ** |
|--------------|----------|----|----|----|----|----|
|              | Stress   | ** | ** | NS | ** | ** |
|              | Interaction| ** | ** | ** | ** | * |

**Significant at 1% level of probability, * Significant at 5% level of probability, NS = Non significant
Yield performances of rice under submergence stress

Table 2. Phenological characters, grain filling rate and maximum grain weight in five rice cultivars as affected by submergence stress at early vegetative stage

| Cultivars       | Submergence stress | Grain filling duration (d) | Days to anthesis (d) | Days to Physiological maturity (d) | Grain filling rate (mg grain\(^{-1}\) d\(^{-1}\)) | Grain weight (mg grain\(^{-1}\)) |
|-----------------|--------------------|---------------------------|----------------------|----------------------------------|-----------------------------------------------|--------------------------------|
| Binadhan-11     | Control            | 34                        | 93                   | 127                              | 0.80                                          | 27.21                          |
|                 | Stress             | 39                        | 106                  | 145                              | 0.64                                          | 25.09                          |
| Binadhan-12     | Control            | 36                        | 111                  | 147                              | 0.44                                          | 15.96                          |
|                 | Stress             | 37                        | 116                  | 153                              | 0.38                                          | 14.11                          |
| BRRI dhan51     | Control            | 40                        | 110                  | 150                              | 0.45                                          | 18.13                          |
|                 | Stress             | 44                        | 124                  | 168                              | 0.36                                          | 15.79                          |
| BRRI Dhan52     | Control            | 42                        | 103                  | 145                              | 0.58                                          | 24.17                          |
|                 | Stress             | 38                        | 107                  | 145                              | 0.60                                          | 22.86                          |
| BRRI Dhan49     | Control            | 45                        | 100                  | 145                              | 0.41                                          | 18.37                          |
|                 | Stress             | 37                        | 111                  | 148                              | 0.27                                          | 10.13                          |

**Significance (F value)**
- Control: **
- Stress: NS
- Interaction: **

** Significant at 1% level of probability, * Significant at 5% level of probability, NS = Non-significant

The average grain-filling rate exhibited a significant variation between the treatments. The range and average of grain filling rate were 0.41–0.80 and 0.54 mg grain\(^{-1}\) day\(^{-1}\) respectively under control treatment. The submergence stress decreased the grain filling rate with the range and mean of 0.27–0.64 and 0.45 mg grain\(^{-1}\) day\(^{-1}\) respectively. Generally, the tolerant cultivars (e.g. Binadhan-11, Binadhan-12, BRRI Dhan51 and BRRI Dhan52) showed comparatively higher grain filling rate compared to the susceptible cultivar (e.g. BRRI Dhan49).

Reduction in yield due to submergence stress

The yield reduction percentage is higher (96.49%) in susceptible cultivar BRRI Dhan49 (Figure 1) than the tolerant cultivars (Binadhan-11, Binadhan-12, BRRI dhan51 and BRRI dhan52) due to submergence stress. Higher the reduction in yield indicates the susceptibility to submergence stress. The lowest reduction (4.4%) was recorded in Binadhan-12. The other cultivars (Binadhan-11, BRRI dhan51 and BRRI dhan52) also showed good performances regarding low reduction in yield (17.42%, 7.55% and 14.50% respectively) which indicates its submergence tolerance ability.

Changes in leaf greenness

The leaf greenness of flag leaves evaluated by SPAD readings is shown in Figure 2. The SPAD reading was about 40–45 in almost all cultivars at 40 days after desubmergence. However, the cultivars exhibited variations in changing pattern of SPAD reading during maturity. At 40 DAD most of the cultivars in stress condition show higher leaf greenness than the control of respective cultivar. After desubmergence, the tolerant cultivars (Binadhan-11, Binadhan-12, BRRI dhan51 and BRRI dhan52) maintain higher leaf greenness for a long time in respect of susceptible cultivar (BRRI dhan49).
Discussion

There was a tremendous effect on grain yield between the rice genotypes due to submergence and the grain yield varies from 0.82 g/pot in BRRI dhan49 to 23.36 g/pot in BRRI dhan49 (Table 2). Yield components that influence the yield of a cultivar under submergence are number of grain/panicle, number of panicle/pot and 1000 grain weight.

In the present experiment, the highest grain yield in control was contributed by BRRI dhan49 (23.36 g/pot) followed by others due to more number of grains/panicle and more number of effective tillers/hill. In submergence stress condition the highest grain yield was contributed by BRRI dhan51 (21.32g/pot) followed by others due to more number of effective tillers/hill which indicates the survival percentage under the submerged condition is maximum as compared to others. The no. of effective tillers/hill was highest (10.00) in BRRI dhan51 and Binadhan-12 followed by Binadhan-11 (9.00) whereas the lowest value of the same was exhibited by BRRI Dhan49 (1.00) which indicate the survival percentage of the cultivar under submergence. Under the submerged condition, the hydrolyzed product of the sucrose by invertase are compensated as a respiratory substrate for the shoot rather than conversion of grain filling materials like starch (Fridman and Zamir, 2003). There is a greater ability to sink tissue to store sugar (Chen and Wang, 2008).

So the number of panicles/hill and 1000-grain weight under the condition of submergence are regarded as the selection criteria for adequate production of grain yield. These earlier finding of the scientists coincide the present investigation. On the other hand lower value of these above yield attributing characters reduced the yield of the susceptible cultivars. The high yield was due to the translocation of the carbohydrate from the stem and leaves after the regeneration and increased biomass production after the submergence (Islam et al., 2010).

The above findings regarding the relationship of grain yield with its attributes were in accordance with the findings of many workers whose reports indicated that the rice yield was positively associated with a number of grains/panicle, and number of panicles per hill (Islam et al., 2010; Li et al., 2012). According to them, the 1000-grain weight may affect rice yield to some extent, but rarely become limiting under most condition.

Therefore, under present investigation importance should be given on a number of effective tillers/pot and a number of grains/panicle and 1000-grain weight to increase the yield potential of rice genotypes under submergence stress.

Most of the cultivars in stress condition show higher leaf greenness than the control of respective cultivar. This is because of the lower age of the stressed plant. The plant under submerged condition had died, but after desubmergence they grow again with young leaves. These young leaves exhibited higher leaf greenness. In case of control, the age of the plant was much more than the submerged plant. These older leaves exhibited lower greenness than the submerged plant.

Conclusion

Increasing submergence duration resulted in decreased tiller number per plant in susceptible cultivars. Submergence tolerant rice cultivars (Binadhan-11, Binadhan-12, BRRI Dhan51 and BRRI Dhan52) had maintained higher tiller number, 1000-grain weight, a higher number of grains per panicle and higher leaf greenness after submergence stress, as compared to susceptible rice cultivar BRRI Dhan49. So these traits can be considered as selection criteria for submergence tolerant rice cultivar.
Yield performances of rice under submergence stress

Acknowledgement

This research was supported by a Research Grant (2017/45/BAU) from Bangladesh Agricultural University Research System, BAU, Mymensingh.

References

Armstrong, W. 1980. Aeration in Higher Plants. Advances in Botanical Research, 7: 225–332. https://doi.org/10.1016/S0065-2296(08)60089-0

BBS, 2016. Yearbook of Agricultural Statistics-2015 (Bangladesh Bureau of Statistics, Statistical Division, Ministry of Planning, Govt. Peoples Republic of Bangladesh, Dhaka.).

Chen, H.J. and Wang, S.J. 2008. Molecular regulation of sink-source transition in rice leaf sheaths during the heading period. Acta Physiologiae Plantarum, 30: 639–649. https://doi.org/10.1007/s11738-008-0160-8

Colmer, T.D. 2003. Long-distance transport of gases in plants: A perspective on internal aeration and radial oxygen loss from roots. Plant, Cell and Environment, 26: 17–36. https://doi.org/10.1046/j.1365-3040.2003.00846.x

Colmer, T.D. and Pedersen, O. 2008a. Oxygen dynamics in submerged rice (Oryza sativa). New Phytologist, 178: 326–334. https://doi.org/10.1111/j.1469-8137.2007.02364.x

Colmer, T.D. and Pedersen, O. 2008b. Underwater photosynthesis and respiration in leaves of submerged wetland plants: Gas films improve CO₂ and O₂ exchange. New Phytologist, 177: 918–926. https://doi.org/10.1111/j.1469-8137.2007.02318.x

Dias, A.S. and Lidon, F.C. 2009. Evaluation of grain filling rate and duration in bread and durum wheat, under heat stress after anthesis. Journal of Agronomy and Crop Science, 195: 137–147. https://doi.org/10.1111/j.1439-037X.2008.00347.x

Drew, M.C. and Lynch, J.M. 1980. Soil anaerobiosis, microorganisms, and root function. Annual Review of Phytopathology, 18: 37–66. https://doi.org/10.1146/annurev.py.18.090180.000345

FAO, 2017. Rice Market Monitor (Food and Agricultural Organization, Rome.).

Fridman, E. and Zamar, D. 2003. Functional divergence of a syntenic invertase gene family in tomato, potato, and arabidopsis. Plant Physiology, 131: 603–609. https://doi.org/10.1104/pp.014431

Islam, M.S., Peng, S., Visperas, R.M., Bhuiya, M.S.U., Hossain, S.A. and Julfiqar, A.W. 2010. Comparative study on yield and yield attributes of hybrid, inbred, and NPT rice genotypes in a tropical irrigated ecosystem. Bangladesh Journal of Agricultural Research, 35: 343–353. https://doi.org/10.3329/bjar.v35i2.5897

Li, X., Yan, W., Agrama, H., Jia, L., Jackson, A., Moldenhauer, K., Yeater, K., McClung, A. and Wu, D. 2012. Unraveling the complex trait of harvest index with association mapping in rice (Oryza sativa L.). PLoS ONE, 7: e29350. https://doi.org/10.1371/journal.pone.0029350

Nishiuchi, S., Yamauchi, T., Takahashi, H., Kotula, L. and Nakazono, M. 2012. Mechanisms for coping with submergence and waterlogging in rice. Rice, 5: 1–15. https://doi.org/10.1186/1939-8433-5-2

Ram, P.C., Singh, B.B., Singh, A.K., Ram, P., Singh, P.N., Singh, H.P., Bounfa, I., Harren, F., Santos, E. and Jackson, M.B. 2002. Submergence tolerance in rainfed lowland rice: Physiological basis and prospects for cultivar improvement through marker-aided breeding. Field Crops Research, 76: 131–152. https://doi.org/10.1016/S0378-4290(02)00035-7