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**AGRONOMY | ORIGINAL ARTICLE**

# **Assessing Deepwater Rice Mutant Populations for Some Morpho-Physiological and Agronomic Traits**

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#### **ARTICLE INFO ABSTRACT**

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Unlike most current rice cultivars, rice produced in the deepwater habitat may live for at least a month in water depths exceeding 50 cm. In this experiment, 30  $M_6$  populations developed by irradiation of *Laksmidigha* rice seeds utilizing 200 Gy dose of gamma rays were assessed based on morphophysiological and agronomic traits. Six mutant populations exhibited noticeably longer leaves, with LD-200-1-1-2-6 being the only mutant population to have noticeably more grain weight hill<sup>-1</sup> than the parent. But none of the mutant populations had significantly wider leaf breadth at any of the three points of the flag leaf and average as well. Six mutant populations LD-200-1-3-3-3, LD-200-1-3-3-4, LD-200-1-3-3-5, LD-200-1-3-2-1, LD-200-1-1-2-1 and LD-200-1-1-2-6 produced significantly higher grain yield hill<sup>-1</sup> which also had significantly longer panicle lengths, internode lengths and heavier 1000-grain weight than the parent. It was discovered through a correlation analysis that grain weight hill<sup>-1</sup> was positively and significantly connected with panicle length and 1000-grain weight, whereas its association with internode distance was positively but not significantly correlated. But the correlation of internode distance with 1000-grain weight was significantly positive. The six mutant populations that significantly increased grain yield hill<sup>-1</sup> compared to the parent also had higher levels of total chlorophyll and chlorophyll "a" in their flag leaves. In anatomical studies of roots, it was found that all the 21 cross sections showed damaged cortex with different degrees except LD-200-1-3-3-7 and LD-200-1-1-2-5. Even the parent grown in Chamber-1 showed minimum cortex damage. Also, the grain yield of these two mutant populations did not differ significantly from the parent although chlorophyll "a" and total chlorophyll contents were much higher.

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# **1. Introduction**

In addition to wheat and maize, rice is one of the most important crops in the world and the mainstay of Bangladesh agriculture. Actuality, a single crop (rice) and one season (*Boro*) account for around 54% of Bangladesh's overall rice output from an area that produces 42% of the country's total rice (AIS 2019). But extensive *Boro* cultivation has been proven to be the cause of environmental degradation, particularly through lifting of huge amount of underground water for irrigation. Moreover, it competes with other Rabi crops like wheat, oilseed, pulse and vegetables due to coincidence of the growing season. Therefore, the government is emphasizing to reduce the area under *Boro* rice. But to feed the ever-growing population and to meet the sustainable development goal (SDG) by 2030, rice

production should be increased by 2-fold. The question is how it can be done? It can be done by increasing rice area in *Aus* and *Aman* seasons. In this study, we are emphasizing on increasing area in *Aman* season. There is a big area of land in haor and bill where hardly *Aus* and *Aman* rices can be grown due to high water depth. In that situation, deep water rice can be a potential crop to withstand such condition.

Deepwater rice refers to rice types that can withstand flooding that is more than 50 cm deep and lasts for at least a month. Such times allow for the development of adventitious roots, which offer water, nutrients, and anchoring. The floating rice among the deepwater rice varieties have a very high elongation capability. When partially submerged, they may develop at rates of 20 to 25 cm/d and reach lengths of up to 7 m in up to 4 m of water

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(Vergara and Chang, 1976, Catling and Puckridge 1988). Rice that floats and conventional talls are two modifications that let rice grow in deeper water. Due to its superior elongation capacity, floating rice can grow in water that is more than 100 cm deep. This implies that when a field of rice is flooded, the plant can maintain some of its foliage above the water due to rapid development in the internodal of the stem.

Prior to the start of a flood, when the plant develops basal tillers, deepwater rice grows on dry soil under rainfed circumstances for a period of two to four months. After being flooded, the plant changes into an emergent macrophyte and spends the final three to five months of its life growing in water. Nodal roots take up nutrients from floodwater, including nitrogen, phosphorus, and other elements. Partial submergence triggers stem elongation, which is caused by cell division and elongation of cells in the intercalary meristem owing to an interaction of the plant hormones and is regulated by two complimentary genes. With increasing water depth, there are more elongated internodes. The majority of Bangladesh's deepwater rice varieties are significant elongators. In extremely deep water (3-4 m), the stem can grow to a height of 5–6 m. At water depths more than 4 meters, deepwater rice typically cannot survive.

Deepwater rice cultivars' growth is greatly influenced by variables such the flooding pattern, micro relief, soil type, etc. The vast majority of deepwater cultivars have high photoperiod sensitivity. Photosensitivity regulates the timing of blooming throughout the flood period, allows the plant to avoid the negative effects of low temperature during the reproductive phase, and often assures crop maturity as soon as the floods have subsided. The ability to grow longer as water levels rise, the development of nodal tillers and roots from the top nodes in the water, and kneeing (an upward bend of the plant's terminal section) which retains the reproductive parts above the water when the flood subsides are all characteristics of deepwater (floating) rice.

The predominant kind of deepwater rice is *Oryza sativa* indica, while *Oryza sativa* japonica variations have been identified in Burma, Bangladesh, and India. It is believed that the deepwater rice, *Oryza sativa*, evolved from the perennial grass, *Oryza rufipogon*, through its annual wild relative, *Oryza nivara*. Asian deepwater rice is thought to have originated in this belt because Bangladesh is located in the heart of the east-west axis where rice may have first been domesticated in Asia. Deepwater rice was a significant crop in Bangladesh up until the late 1960s, taking up roughly 2.09 million hectares (21% of the country's total rice area). The introduction of high yielding varieties (HYV) under irrigation in deepwater rice fields during the dry season (*Boro*) led to a subsequent reduction in the cropping area to around 0.85 million hectares. million acres are currently fallow. Deep water rice (DWR) is now grown in *haor* and *beel* regions like Sunamganj, Sylhet, Habiganj, B. Baria, Faridpur, Gopalganj, and Pabna over an area of 0.48 million ha (BBS 2009). Despite DWR's limited yield and small-scale cultivation, care should be paid to unlocking its full production potential.

Given the foregoing, we examined thirty high yielding mutant lines of DWR for morpho-physiological, anatomical traits and their role in determining the grain yield, agronomic traits and their interrelationships.

# **2. Materials and Methods**

## **2.1. Site description**

This research was conducted in Deepwater rice Screening Tank (DWRST) of Plant Breeding Division, Bangladesh Institute of Nuclear Agriculture (BINA), Mymensingh during July to December 2017 (monsoon rice growing season).

# **2.2. Experimental setup**

Thirty M<sup>6</sup> populations (Table 1) of deepwater rice developed by Plant Breeding Division of BINA were used in this experiment. The local deepwater rice cultivar *Laksmidigha* seeds were first exposed to a 200 Gy dose of gamma radiation before being selected for plant height, stem elongation rate, and the number of grains per panicle. This process created the M<sup>6</sup> populations. The seeds of *Laksmidigha* were irradiated in 2011 from the <sup>60</sup>Co source of Institute of Food and Radiation Biology (IFRB), Savar. The pedigrees of the mutant populations are preented in Table 1. This experiment followed non replicated progeny-rows. A row of 3 m length comprised a progeny. Ten progenies along with a row of parent cultivar were accommodated in each of the three chambers of deep-water rice screening tank (DWRST).

# **2.3. Crop husbandry**

The experimental land was puddled by spading the soil followed by addition of water, removal of weeds and stubbles and finally by laddering. For rapid germination, the seeds were placed in damp gunny sacks for 48 hours after soaking in water for 24 hours. The sprouted seeds were sown on seedbed on at 31 separate beds of required sizes. On July 7, 2017, one seedling hill<sup>-1</sup> was moved into a row that was 20 cm apart at a spacing of 15 cm. Out of the three chambers of the tank, in Chamber-1 ten mutant populations with pedigrees LD-200-1-1-2 to LD-200-1-1- 10 were transplanted, in Chamber-2 ten other mutant populations with pedigrees LD-200-1-2-1 to LD-200-1-2- 10 and the remaining 10 mutant populations with pedigrees LD-200-1-3-1 to LD-200-1-3-10 in Chamber-3 were transplanted. In each of the three chambers, a row of the parent *Laksmidigha* was also transplanted. Fertilizers were administered @ 150 kg of urea, 80 kg of TSP, 60 kilogram of mop, 20 kg ha<sup>-1</sup> of gypsum, and 5 tons ha<sup>-1</sup> of farmyard manure. The FYM was applied 15 days before final land preparation. At the time of the last stage of land preparation, the complete TSP, MoP, and Gypsum were applied. As a top dressing, urea was administered in three equal portions. Following the establishment of the seedlings, the first installment was administered. The second and third installments followed 20–25 and 30-35 days later. After 60 days of transplanting, water depth was raised of the tank during 28 July to 27 September 2017 at every alternate day using the inlet pipe (Table 2).

SI. No.	Pedigree of the mutant populations	SI. No.	Pedigree of the mutant populations
	LD-200-1-1-2-1	8	$LD-200-1-1-2-8$
	LD-200-1-1-2-2	9	$LD-200-1-1-2-9$
3	$LD-200-1-1-2-3$	10	LD-200-1-1-2-10
4	LD-200-1-1-2-4	11	LD-200-1-3-2-1
5	$LD-200-1-1-2-5$	12	$LD-200-1-3-2-2$
6	LD-200-1-1-2-6	13	$LD-200-1-3-2-3$
	LD-200-1-1-2-7	14	$LD-200-1-3-2-4$
15	LD-200-1-3-2-5	23	$LD-200-1-3-3-3$
16	LD-200-1-3-2-6	24	$LD-200-1-3-3-4$
17	LD-200-1-3-2-7	25	LD-200-1-3-3-5
18	LD-200-1-3-2-8	26	$LD-200-1-3-3-6$
19	$LD-200-1-3-2-9$	27	$LD-200-1-3-3-7$
20	LD-200-1-3-2-10	28	$LD-200-1-3-3-8$
21	LD-200-1-3-3-1	29	LD-200-1-3-3-9
22	LD-200-1-3-3-2	30	LD-200-1-3-3-10

**Table 1.** Pedigree of the deepwater rice mutant populations

**Table 2.** Gradual increase of water depth in the three chambers of Deepwater Rice Screening Tank

Date of water depth measurement	<b>Chamber-1 water depth</b>	<b>Chamber-2 water depth</b>	<b>Chamber-3 water depth</b>		
	(cm)	(cm)	(c <sub>m</sub> )		
28/7/2017	95	97	100		
31/07/2017	104	105	112		
06/08/2017	108	108	118		
23/08/2017	129	150	177		
17/07/2017	112.5	125.5	144.5		
20/07/2017	112.5	130	169		
29/08/2017	125	150	183		
30/08/2017	125	155	185		
27/09/2017	179	193	183		

Two hand weeding were made before top dressing of urea at 2<sup>nd</sup> and 3<sup>rd</sup> installments along with application of required irrigation water. Appropriate control measures for insect-pest and diseases were also taken. At maturity, harvesting was completed. Different lines reach different stages of development at various periods. The grain was only to be harvested after 80% of it became golden yellow.

## **2.4. Sampling and measurements**

#### *2.4.1. Morpho-physiological data*

Leaf length and breadth of five randomly selected flag leaves of five hills per population was measured. Leaf length was measured from the base of the flag leaf to its tip and breadth from base, middle and tip.

#### *2.4.2. Estimation of chlorophyll content in leaf*

The method of (Shabala et al. 1988) was followed with some modifications. One leaflet was taken from the uppermost fully expanded compound leaf of the main stem and was cut into pieces leaving the mid rib. The leaflets were cut between 100-120 days after sowing the plants. From the pieces 0.1g leaf was dipped into 80% ethanol and kept in a dark place for 2 weeks at room temperature. Readings were recorded at 645 and 663 nm wavelengths from a spectrophotometer. Finally, chlorophyll 'a' chlorophyll 'b' and total chlorophyll were estimated following (Yoshida et al. 1976).

Total chlorophyll =  $(20.2 \times D645 + 8.20 \times D663) \times DF$ **Where** 

D645=Absorbance at 645 nm wavelength

D663= Absorbance at 663 nm wavelength

#### *2.4.3. Root anatomy study of adventitious roots*

Adventitious roots of 30 M6 populations and the parent were collected and preserved in FAA solution. Small thin layer of root was cut from the sample then taking in the slide and added little amount of glycerin so that root would not get dry. Then root anatomy was studied by observing arenchyma in the root cell under a microscope. After that some picture was taken through electronic microscope for future study.

#### **2.5. Data collection**

Data on 13 characters were recorded. Among the studied characters, plant height, no. of primary, secondary and effective tillers, panicle length, no. of internode and internode distance were recorded in the field and other characters grain weight per hill ,1000-grain weight, no. of filled and unfilled grains per panicle were recorded in the laboratory after harvesting. The five hills from the population that were randomly chosen to collect these statistics.

## **2.6. Statistical analysis**

All the collected data was collected and tabulated, and the statistical analysis of those data was performed through Microsoft Excel.

# **3. Results**

#### **3.1. Trait-wise mean performance of the morphophysiological parameters of 30 deepwater rice mutant populations along with the parent**

The mean performances of morpho-physiological traits of 30 deepwater rice mutant populations and their parent are shown in Table 3. The mutant populations' flag leaf lengths varied from 20.48 cm to 45.90 cm, with LD-200-1- 1-2-5 having the smallest length and LD-200-1-3-2-7 having the greatest (Table 3). Most of the mutant populations had more intra population variation than the parent *Laksmidigha* as indicated by higher standard error. Only 10 mutant populations showed lower intra population variation. Seven mutant populations had significantly longer flag leaf length whereas four had shorter length than the parent.

Breadth of flag leaf at base of the mutant populations fluctuated from 0.86 cm to 1.76 cm with LD-200-1-3-2-6 being the narrowest and LD-200-1-3-3-5 the widest. All the mutant populations had less intra population variation for breadth of flag leaf base than the parent *Laksmidigha* except three. Seventeen mutant populations had significantly narrower flag leaf base than the parent. But no one had broader flag leaf base than the parent. Breadth of flag leaf at middle point of the mutant populations varied from 1.16 cm to 1.78 cm with LD-200-1-3-2-6 being the narrowest and LD-200-1-1-2-1 the widest. Ten mutant populations had less intra population variation for breadth of flag leaf at middle point than the parent *Laksmidigha*. Twenty-seven mutant populations had significantly narrower breadth of flag leaf at the middle point than the parent. But no one had broader flag leaf at middle point than the parent.

Breadth of flag leaf at the tip of the mutant populations is ranged from 0.3 cm to 1.1cm with LD-200-1-3-2-1 being the narrowest and LD-200-1-1-2-3 the widest. Nine mutant populations had less intra population variation for breadth of flag leaf at the tip than the parent *Laksmidigha*. Twentytwo mutant populations had significantly narrower and only one had broader breadth of flag leaf at middle than the parent. The average breadth of flag leaf ranged from 0.80 cm to 1.42 cm with LD-200-1-3-2-6 being the narrowest and LD-200-1-1-2-1 the widest (Table 3). Seven mutant populations had less intra population variation for average breadth of flag leaf than the parent *Laksmidigha*. Twenty-two mutant populations had significantly narrower average breadth of flag leaf than the parent (Table 3).

Chlorophyll ''a'' content of the mutant populations ranged from 12.8 to 33.8 mg/gfwt with LD-200-1-1-2-7 being the lowest and LD-200-1-3-2-1 the highest. Compared to the parent, all of the mutant populations exhibited chlorophyll "a" content in the leaves that varied among populations either more or equally. Chlorophyll ''b'' content of the mutant populations ranged from 5.6 to 30.0 mg/gfwt with LD-200-1-1-2-4 being the lowest and LD-200-1-3-2-1 the

highest. All the mutant populations had either higher or equal intra population variation for chlorophyll ''b'' content in leaf. Total chlorophyll content of the mutant populations ranged from 19.7to 63.7 mg/gfwt with LD-200-1-1-2-7 being the lowest and LD-200-1-3-2-1 the highest. For the overall chlorophyll content in the leaf, all the mutant populations either displayed more or comparable intra population variance (Table 3).

#### **3.2. Trait-wise agronomic mean performance of 30 deepwater rice mutant populations along with the parent**

The mean agronomic performances of 30 deepwater rice mutant populations and their parent are shown in Table 2. The mutant populations' plants ranged in height from 293.8 cm to 341.4 cm, with LD-200-1-3-3-10 being the smallest and LD-200-1-3-2-2 being the highest (Table 4). Most of the mutant populations had more intra population variations than the parent *Laksmidigha* as indicated by higher standard errors. Only 13 mutant populations showed lower intra population variation. Four mutant populations had longer plant height although with no significant difference but 19 produced shortest plant than the parent. Number of primary tillers of the mutant populations ranged from 1.4 to 5.4 with LD-200-1-3-3-2 and LD-200-1-3-3-8 being the lowest and LD-200-1-3-3-4 the highest. Most of the mutant populations had lower intra population variations than the parent *Laksmidigha* as indicated by lower standard errors. Only 12 mutant populations showed higher or equal intra population variations. No mutant populations had significantly higher primary tiller whereas 16 had lower number of primary tiller than the parent.

Nodal tiller of the mutant populations ranged from 1.4 to 14.0 with LD-200-1-3-2-8 being the lowest and LD-200-1- 3-2-6 the highest. Twenty mutant populations had lower intra population variations than the parent *Laksmidigha* as indicated by lower standard errors. Only ten mutant populations showed higher intra population variations. No mutant population showed a noticeably larger number of nodal tillers than the parental population. Effective tiller of the mutant populations ranged from 1.8 to 13.0 with LD-200-1-3-2-8, LD-200-1-3-2-10 and LD-200-1-1-2-10 being the lowest and LD-200-1-3-2-6 the highest. Twenty-one mutant populations had lower intra population variations than the parent *Laksmidigha* as indicated by lower standard errors. Only ten mutant populations showed higher or equal intra population variations. There were no substantially more effective tillers in any of the mutant groups than in the parent population. Panicle length of the mutant populations varied from 25.0 cm to 32.8 cm with LD-200-1-3-3-1 being the shortest and LD-200-1-3-2-10 the longest. Eight of the mutant populations had lower intra population variations than the parent *Laksmidigha* as indicated by lower standard errors. The other mutant populations showed either higher or equal intra population variations. All mutant populations had noticeably longer panicles than the parent, which was a notable finding. Internode number of the mutant populations ranged from 11.4 to 20.2 with LD-200-1-3-2-3 being the lowest and LD-200-1-3-3-7 the highest. Fourteen of the mutant populations had lower intra population variations than the parent *Laksmidigha* as indicated by lower standard errors. The other mutant populations showed either higher or equal intra population variations. It's interesting to see that none of the mutant populations had noticeably more internodes than the parent population. The internode distance of the mutant populations ranged from 15.4 to 30.2 with LD-200-1-3-2-3 being the lowest and LD-200-1- 3-2-10 the highest. Only one mutant had lower intra population variation than the parent *Laksmidigha* as indicated by lower standard error. The other mutant populations had higher intra population variations in internode distance than the parent. Twenty-four mutant populations had significantly longer internode distance than the parent. Filled grains panicle<sup>-1</sup> of the mutant populations ranged from 111.8 to 230.4 with LD-200-1-3- 2-3 being the lowest and LD-200-1-3-3-10 the highest. Seven of the mutant populations had lower intra population variations than the parent *Laksmidigha* as indicated by lower standard errors. Twenty-three mutant populations showed higher or equal intra population variations. Four of the mutant populations had fewer filled grains panicle<sup>-1</sup> than the parent, whereas 13 had considerably greater filled grains panicle<sup>-1</sup>.

Unfilled grains panicle-1 of the mutant populations ranged from 35.4 to 138.6 with LD-200-1-3-2-6 being the lowest and LD-200-1-3-2-8 the highest. Twelve of the mutant populations had lower intra population variations than the parent *Laksmidigha* as indicated by lower standard errors. The other mutant populations showed higher intra population variations. Eleven mutant populations had significantly higher unfilled grains panicle<sup>-1</sup> than the parent and only one mutant had significantly lower unfilled grains/panicle. The grain weight hill<sup>-1</sup> of the mutant populations ranged from 4.6 to 18.1 g with LD-200-1-1-2- 8 being the lowest and LD-200-1-3-3-3 the highest (Table 4). Thirteen of the mutant populations had lower intra population variations than the parent *Laksmidigha* as indicated by lower standard errors. The other mutant populations showed higher intra population variations. Only three mutant populations exhibited grain weight hill<sup>-1</sup> that was considerably greater than the parent. 1000-grain weight hill<sup>-1</sup> of the mutant populations ranged from 15.7 to 22.0 g with LD-200-1-3-3-8 being the lowest and the highest LD-200-1-3-3-10 (Table 4). Fifteen mutant populations had lower intra population variations than the parent *Laksmidigha* as indicated by lower standard errors. The other mutant populations showed higher or equal intra population variations. Twenty-five mutant populations had significantly higher thousand-grain weight than the parent (Table 4).

## **3.3. Phenotypic correlation coefficients of grain yield with other agronomic traits of 30 deepwater rice mutant populations**

It is evident that grain weight hill<sup>-1</sup> has significant positive correlations with all other agronomic traits except number of internode and unfilled grains panicle<sup>-1</sup> (Table 5). Conversely, unfilled grains panicle<sup>-1</sup> revealed a negative significant association whereas the no. of internodes indicated a negative not significant correlation. Number of nodal tillers, effective tiller, panicle length, no. of filled grains panicle<sup>-1</sup> and 1000-grain weight showed significant positive correlations with grain weight hill<sup>-1</sup>. Thousand grain weight showed positive correlations with panicle length, number of internodes, internode distance, no. of filled and unfilled grains panicle<sup>-1</sup>. Of which correlations with internode distance and number of filled grains panicle-1 were significant.

No. of unfilled grains panicle<sup>-1</sup> showed significant positive correlations with panicle length only and not significant positive with number of internodes, internode distance and number of unfilled grains panicle<sup>-1</sup>. Interestingly, no. of unfilled grains panicle<sup>-1</sup> had significant negative correlation with plant height. Number of filled grains panicle-1 showed significant positive correlations with number of primary tillers, nodal tiller and panicle length. Of which the correlation with panicle length was the highest of all. In contrast, its correlation with plant height was the highest but negative. Only the distance between internodes and the length of the panicle exhibited a positive association, but the number of internodes exhibited a significant positive correlation with the height of the plant but not with the number of primary tillers. While there was a substantial negative link between panicle length and plant height, there was no such correlation with the number of primary, secondary, and effective tillers. In contrast, effective tiller showed significant positive correlations with plant height, no. of primary and nodal tillers. Nodal tiller showed positive significant correlations with plant height and primary tiller but primary tiller showed not significant positive correlation with plant height (Table 5).

#### **3.4. Adventitious root cross section of the deepwater rice mutant populations**

The cross sections of the adventitious root of some of the deepwater rice mutant populations along with the parent *Laksmidigha* are shown in Figure 1. It was revealed that the cortex of most of the mutant populations were damaged forming arenchyma. Less arenchyma were found in the mutant populations LD-200-1-3-3-6 and LD-200-1-3-3-8 and the parent in Chamber-1 [Figure 1(f), 1(h) & 1 (w)]. But the other mutant populations had higher number of arenchyma.

# **4. Discussion**

#### **4.1. Trait-wise mean performance of the morphophysiological parameters of 30 deepwater rice mutant populations along with the parent**

Genetic diversity is essential for introducing novel features into breeding programs since plant breeding depends on it. However, different methods can be used to increase the genetic diversity in a crop species when a particular genetic feature is not readily available to be crossed into breeding materials. Induced mutation is an easy, proven and robust method for inducing variations in crop plants (Lagoda and Forster 2013, Azad et al. 2010). There are many indigenous cultivars of deepwater rice in Bangladesh but with very low yield. The conventional/cross breeding mostly failed to develop higher yielding variety of deepwater rice. Therefore, attempt was taken to develop higher yielding varieties of deepwater rice using induced mutation breeding. The seeds of the local deepwater rice variety *Laksmidigha* were given a 200 Gy dosage of gamma radiation in 2011, and then they were chosen based on the plants' height, panicle length, grain production per plant, and 1000-grain weight. Thirty mutant populations were obtained. These mutant populations have been assessed in this study for some morpho-physiolocal and agronomic traits.



**Figure 1.** Adventitious root cross section of the deepwater rice mutant populations. a) LD-200-1-3-3-1 mutant, b) LD-200- 1-3-3-2 mutant, c) LD-200- 1-3-3-3 mutant, d) LD-200- 1-3-3-4 mutant, e) LD-200- 1-3-3-5 mutant, f) LD-200- 1-3-3-6 mutant, g) LD-200- 1-3-3-7 mutant, h) LD-200- 1-3-3-8 mutant, i) LD-200- 1-3-2-1 mutant, j) LD-200- 1-3-2-2 mutant, k) LD-200- 1-3-2-3 mutant, l) LD-200- 1- 3-2-4 mutant, m) LD-200- 1-3-2-5 mutant, n) LD-200- 1-3- 2-6 mutant, o) LD-200- 1-3-2-7 mutant, p) Parent (in Chamber-3), q) LD-200- 1-1-2-1 mutant, r) LD-200- 1-1-2- 2 mutant, s) LD-200- 1-1-2-3 mutant, t) LD-200- 1-1-2-5 mutant, u) LD-200- 1-1-2-7 mutant, v) LD-200- 1-1-2-9 mutant, w) parent *Laksmidigha* ( in Chamber-1)

There were intra and inter population variations in flag leaf length, leaf breadth (base, middle point, tip and average), chlorophyll "a", chlorophyll "b" and total chlorophyll contents. Nine mutant populations had lower intra population variations for flag leaf length, 24 for leaf breadth at base, 11 for leaf breadth at middle point, 9 for leaf breadth at tip and 8 for average leaf breadth than the parent which indicated these populations already attained homozygosity for these morpho-physiological traits but the remainders need further selection. Six mutant populations had significantly longer leaf length than the parent of which one mutant population LD-200-1-1-2-6 had significantly higher grain weight/hill than the parent. But none of the mutant population had significantly wider leaf breadth at any of the three points of the flag leaf and average as well.

#### **4.2. Trait-wise performance of 30 deepwater rice mutant populations along with the parent**

There were intra and inter population variations in plant height indicated scope of selections within and among populations. Thirteen mutant populations with fewer intrapopulation differences for plant height than the parent populations suggested that these populations had already achieved homozygosity and were therefore not subject to additional selection based on this feature. These results are in line with that of (Nafis et al. 2018). Similar intra and inter population variations were also observed for no. of primary tiller, no. of nodal tiller, effective tiller, panicle

length, no. of internode, internode distance, filled and unfilled grains panicle<sup>-1</sup>, grain weight hill<sup>-1</sup> and 1000-grain weight.

Eleven mutant populations had higher grain weight hill-1 than the parent. Of which only six, LD-200-1-3-3-3, LD-200-1-3-3-4, LD-200-1-3-3-5, LD-200-1-3-2-1, LD-200-1- 1-2-1 and LD-200-1-1-2-6 exhibited significant differences with the parent. These six mutant populations also had significantly longer panicle lengths, internode distances and heavier 1000-grain weight than the parent. This means for selecting deepwater rice mutant with higher grain yield, longer panicle length, longer internode distance and heavier 1000-grain weight are important criteria. These results were partially supported by the correlation study. The results of a correlation analysis revealed a positive and substantial association between grain weight/hill with panicle length, 1000-grain weight while its correlation with internode distance was positive but not significant. But the correlation of internode distance with 1000-grain weight was significantly positive.

The six mutant populations that generated significantly higher grain weight hill<sup>-1</sup> than the parent also had higher amount of chlorophyll "a" and total chlorophyll in their flag leaf.

#### **4.3. Adventitious root cross section of the deepwater rice mutant populations**

When deepwater rice is flooded, adventitious roots form from the nodes to supply water and nutrients to the plant's newly growing top sections. These roots, which deepwater rice initiates as part of typical plant growth, are shootborne roots. The root initials evolve into root primordia as the plant grows, and these primordia have all the characteristics of primary or lateral roots, including the link to the vasculature. But without submergence, the root primordia won't break through the nodal epidermis (Suge 1985, Bleecker 1987). The low level of oxygen in flooded environments leads to the production of anaerobically induced polypeptides (ANPs), and ACC, the precursor to ethylene biosynthesis. This signal was transmitted from the plant's base to its aerial portion in the shoot. ACCoxidase and oxygen-containing ACC combine to form ethylene. Below the root tip, ethylene causes the creation of aerenchyma, or the gap between cells. The gas exchanges depend heavily on this tissue (Mulyani 2006). Lysigenous or schizogenous processes may be used to create the aerenchyma. A planned cell death led to the lysigenous process, whereas cell separation during tissue development caused the schizogenous process to emerge (Drew et al. 2000, Evans 2003).

In this study, adventitious root cross sections of 21 mutant populations along with two cross sections of the parent have been shown in Figure 1. All the cross sections showed damaged cortex with different degrees except LD-200-1-3-3-7 (Fig. 2g) and LD-200-1-1-2-5 (Fig. 1t). Cross sections of these two mutant populations showed no cortex damage. Even the parent grown in Chamber-1 showed minimum cortex damage (Fig. 1w).

	Leaf length (cm)			Leaf Breadth (cm)	Chlorophyll content (mg g <sup>-1</sup> fresh weight)			
<b>Mutant populations</b>		<b>Base</b>	<b>Middle</b>	<b>Tip</b>	Average	<b>Chlorophyll "a"</b>	<b>Chlorophyll "b"</b>	<b>Total Chlorophyll</b>
LD-200-1-3-3-1	$37.84$ ns ± 3.52	$1.06** \pm 0.02$	$1.42** \pm 0.05$	$0.84$ ns ± 0.03	$1.1*** \pm 0.02$	$20.5 \pm 0.56$	$13.4 \pm 0.53$	$33.9 \pm 1.03$
LD-200-1-3-3-2	$36.28$ ns ± 2.56	$1.0** \pm 0.06$	$1.48* \pm 0.09$	$0.68^* \pm 0.06$	$1.05** \pm 0.05$	$25.7 \pm 0.56$	$12.8 \pm 0.53$	$38.5 \pm 1.03$
LD-200-1-3-3-3	$39.40$ ns ± 3.37	$0.98^{**} \pm 0.05$	$1.08^{**} \pm 0.05$	$0.72 * \pm 0.06$	$0.92** \pm 0.05$	$24.7 \pm 0.56$	$12.8 \pm 0.53$	$37.5 \pm 1.03$
LD-200-1-3-3-4	$33.11$ ns ± 3.68	$1.36$ ns ± 0.09	$1.54** \pm 0.04$	$0.86$ ns ± 0.03	$1.25$ ns $\pm 0.05$	$23.3 \pm 0.56$	$11.9 \pm 0.53$	$35.3 \pm 1.03$
LD-200-1-3-3-5	$27.64$ ns ± 2.00	$1.76$ ns $\pm$ 0.05	$1.34** \pm 0.06$	$0.6** \pm 0.04$	$1.23$ ns ± 0.17	$22.4 \pm 0.56$	$11.4 \pm 0.53$	$33.8 \pm 1.03$
LD-200-1-3-3-6	$28.14$ ns ± 1.79	$1.12** \pm 0.05$	$1.44** \pm 0.06$	$0.62** \pm 0.05$	$1.06** \pm 0.04$	$26.9 \pm 0.56$	$16.7 \pm 0.53$	$43.7 \pm 1.03$
LD-200-1-3-3-7	$36.88$ ns ± 3.03	$1.2$ ns ± 0.09	$1.56** \pm 0.03$	$0.64 *_{\pm} 0.06$	$1.13 * \pm 0.05$	$25.8 \pm 0.56$	$13.6 \pm 0.53$	$39.5 \pm 1.03$
LD-200-1-3-3-8	$36.44* \pm 2.50$	$1.1*** \pm 0.04$	$1.42** \pm 0.01$	$0.66* \pm 0.08$	$1.06** \pm 0.02$	$21.7 \pm 0.56$	$11.2 \pm 0.53$	$33.1 \pm 1.03$
LD-200-1-3-3-9	$36.90$ ns $\pm 3.23$	$1.04* \pm 0.08$	$1.42** \pm 0.07$	$0.64 *_{\pm} 0.08$	$1.03** \pm 0.06$	$23.3 \pm 0.56$	$14.6 \pm 0.53$	$37.9 \pm 1.03$
LD-200-1-3-3-10	$29.06$ ns ± 2.59	$1.2^* \pm 0.04$	$1.44** \pm 0.02$	$0.54$ ** $\pm$ 0.06	$1.06** \pm 0.03$	$23.3 \pm 0.56$	$12.4 \pm 0.53$	$35.7 \pm 1.03$
LD-200-1-3-2-1	$31.84$ ns ± 1.11	$1.08** \pm 0.03$	$1.2** \pm 0.02$	$0.3** \pm 0.02$	$0.86** \pm 0.03$	$33.8 \pm 1.41$	$30.0 \pm 1.82$	$63.7 \pm 3.04$
LD-200-1-3-2-2	$34.86$ ns ± 2.68	$0.98** \pm 0.01$	$1.36** \pm 0.03$	$0.36** \pm 0.03$	$0.9** \pm 0.02$	$26.0 \pm 1.41$	$13.3 \pm 1.82$	$39.4 \pm 3.04$
LD-200-1-3-2-3	$36.00$ ns ± 2.52	$0.88$ ** $\pm 0.07$	$1.22$ ** $\pm$ 0.05	$0.38** \pm 0.03$	$0.82^{**} \pm 0.03$	$24.1 \pm 1.41$	$11.9 \pm 1.82$	$36.1 \pm 3.04$
LD-200-1-3-2-4	$44.26** \pm 2.63$	$1.04** \pm 0.04$	$1.24$ ** $\pm$ 0.04	$0.34^{**} \pm 0.02$	$0.87$ $^{**}\pm$ $0.01$	$25.3 \pm 1.41$	$19.1 \pm 1.82$	$44.5 \pm 3.04$
LD-200-1-3-2-5	$42.04** \pm 1.86$	$0.86^{\ast\ast} \pm 0.02$	$1.18** \pm 0.05$	$0.42** \pm 0.04$	$0.82^{**} \pm 0.03$	$16.6 \pm 1.41$	$10.4 \pm 1.82$	$27.1 \pm 3.04$
LD-200-1-3-2-6	$37.80$ ns ± 3.53	$0.86 *_{\pm} 0.01$	$1.16* \pm 0.01$	$0.4** \pm 0.06$	$0.80* \pm 0.01$	$18.7 \pm 1.41$	$12.9 \pm 1.82$	$31.7 \pm 3.04$
LD-200-1-3-2-7	$45.90** \pm 0.84$	$0.86* \pm 0.01$	$1.36** \pm 0.08$	$0.54** \pm 0.05$	$0.92** \pm 0.08$	$25.2 \pm 1.41$	$20.4 \pm 1.82$	$45.6 \pm 3.04$
LD-200-1-3-2-8	$40.84$ $^{\star}$ $\pm$ $2.37$	$1.22$ ns ± 0.05	$1.36** \pm 0.04$	$0.58** \pm 0.03$	$1.05** \pm 0.02$	$21.1 \pm 1.41$	$13.8 \pm 1.82$	$34.9 \pm 3.04$
LD-200-1-3-2-9	$39.04** \pm 1.43$	$1.04** \pm 0.04$	$1.22** \pm 0.05$	$0.54* \pm 0.09$	$0.93** \pm 0.04$	$26.1 \pm 1.41$	$12.3 \pm 1.82$	$38.4 \pm 3.04$
LD-200-1-3-2-10	$32.40$ ns $\pm$ 1.22	$1.06** \pm 0.03$	$1.22** \pm 0.03$	$0.36** \pm 0.03$	$0.88^{\ast\ast} \pm 0.03$	$24.8 \pm 1.41$	$21.3 \pm 1.82$	$46.1 \pm 3.04$
LD-200-1-1-2-1	$27.62$ ns ± 1.64	$1.46$ ns $\pm$ 0.03	$1.78$ ns $\pm 0.07$	$1.02$ ns $\pm 0.12$	$1.42$ ns $\pm 0.04$	$25.0 \pm 1.84$	$18.8 \pm 2.01$	$43.9 \pm 4.00$
LD-200-1-1-2-2	$28.56$ ns ± 2.20	$1.4$ ns ± 0.06	$1.7$ ** $\pm$ 0.02	$0.96$ ns ± 0.02	$1.35$ ns ± 0.03	$15.2 \pm 1.84$	$6.0 \pm 2.01$	$21.2 \pm 4.00$
LD-200-1-1-2-3	$27.56 * ± 1.12$	$1.36$ ns $\pm$ 0.07	$1.78$ ns $\pm 0.09$	$1.1* \pm 0.05$	$1.41$ ns ± 0.04	$19.9 \pm 1.84$	$8.2 \pm 2.01$	$28.1 \pm 4.00$
LD-200-1-1-2-4	$27.42$ ns ± 2.41	$1.44$ ns ± 0.06	$1.74$ ns $\pm 0.07$	$1.04$ ns ± 0.06	$1.40$ ns ± 0.01	$15.3 \pm 1.84$	$5.6 \pm 2.01$	$20.9 \pm 4.00$
LD-200-1-1-2-5	$20.48* \pm 2.43$	$1.06** \pm 0.03$	$1.38** \pm 0.01$	$1.0$ ns $\pm 0.09$	$1.14*** \pm 0.03$	$27.4 \pm 1.84$	$22.2 \pm 2.01$	$49.6 \pm 4.00$
LD-200-1-1-2-6	$39.80** \pm 0.62$	$1.48$ ns ± 0.04	$1.7** \pm 0.02$	$0.8$ ns ± 0.06	$1.32$ ns $\pm 0.03$	$23.0 \pm 1.84$	$17.0 \pm 2.01$	$40.0 \pm 4.00$
LD-200-1-1-2-7	24.16 ** $\pm$ 0.87	$1.38$ ns ± 0.07	$1.54** \pm 0.02$	$0.62^* \pm 0.06$	$1.18** \pm 0.02$	$12.1 \pm 1.84$	$7.6 \pm 2.01$	$19.7 \pm 4.00$
LD-200-1-1-2-8	$27.34* \pm 1.00$	$1.34$ ns ± 0.04	$1.5$ ** $\pm$ 0.04	$0.64^{\ast} \pm 0.06$	$1.16$ ** $\pm$ 0.03	$19.8 \pm 1.84$	$9.7 \pm 2.01$	$29.6 \pm 4.00$
LD-200-1-1-2-9	$28.00$ ns ± 1.25	$1.18$ ns ± 0.07	$1.4$ ** $\pm$ 0.04	$0.62^* \pm 0.07$	$1.06** \pm 0.03$	$16.7 \pm 1.84$	$9.0 \pm 2.01$	$25.8 \pm 4.00$
LD-200-1-1-2-10	$29.70$ ns ± 2.74	$1.36$ ns ± 0.08	$1.44** \pm 0.04$	$0.6 * \pm 0.06$	$1.13** \pm 0.04$	$31.8 \pm 1.84$	$26.1 \pm 2.01$	$57.9 \pm 4.00$
Laksmidigha (P)	$30.52 \pm 1.60$	$1.36 \pm 0.08$	$1.84 \pm 0.04$	$0.92 \pm +0.04$	$1.37 \pm 0.03$	$20.5 \pm 0.56$	$13.4 \pm 0.53$	$33.9 \pm 1.03$
Range	24.16 to45.90	0.86 to 1.76	1.08 to 1.78	0.36 to 1.0	0.80 to 1.42	12.1 to 33.8	5.6 to 26.1	19.7 to 63.7

**Table 3.** Means of different morpho-physiological parameters of 30 deepwater rice mutant populations along with the parent

\*, \*\*- denote significant at P>0.05 and P>0.01 levels of probability at 4 df according to student `t' test, respectively; ns- denote not significant; P- denotes Parent



# **Table 4.** Means agronomic traits of 30 deepwater rice mutant populations along with the parent

\*, \*\*- denote significant at P>0.05 and P>0.01 levels of probability at 4 df according to student `t' test, respectively; ns- denote not significant; P- denotes Parent

It has been reported in a study that the more the depth of submergence the more will be the cortex damage or aerenchyma formation and at 5 cm depth the root cells remain as those in favorable condition (Nurrahma et al.

2017). This phenomenon may explain no/low aerenchyma formation by two mutant populations along with the parent.

**Table 5.** Correlation coefficient between yield and yield attributing traits of 30 deepwater rice mutant populations

<b>Parameters</b>	<b>PH</b>	<b>PT</b>	<b>ND</b>	ET	<b>PL</b>	<b>IN</b>	IL	FG	<b>UG</b>	<b>WTG</b>	<b>GW</b>
Plant height (cm)											
Primary tiller (no.)	0.418	1									
Nodal tiller (no.)	$0.611*$	$0.8***$									
Effective tiller (no.)	$0.649*$	$0.81***$	$0.99**$								
Panicle length (cm)	$-0.72*$	$-0.4$	$-0.539$	$-0.575$	-1						
No. of internode	$0.679*$	0.074	$-0.04$	$-0.069$	$-0.203$						
Internode distance (cm)	$-0.354$	$-0.279$	$-0.367$	$-0.378$	0.527	$-0.151$	1				
No. of filled grains panicle <sup>-1</sup>	$-0.458$	0.029	0.011	$-0.072$	0.441	$-0.226$	$-0.098$	1			
No. of unfilled grains/panicle	$-0.69*$	$-0.468$	$-0.286$	$-0.335$	$0.638*$	0.096	0.357	0.319	1		
1000 grain weight (g)	$-0.377$	$-0.317$	$-0.372$	$-0.372$	0.283	0.004	$0.635*$	$0.638*$	0.536	1	
Grain weight hill- $(q)$	0.348	0.428	$0.602*$	$0.687*$	$0.721*$	$-0.057$	0.167	$0.81**$	$-0.62*$	$0.78**$	

\*indicates significant at 0.001% probability, \*\*indicates significant at 0.01% probability, \*\*\*indicates significant at 0.5% probability; PH: plant height (cm), PT: number of primary tillers, NT: number of nodal tillers, ET: number of effective tillers, PL: panicle length (cm), IN: number of internodes, internode length (cm), FG: number of filled grains/panicle, UG: number of unfilled grains/panicle, weight of 1000 grains (g), GY: grain weight (g)

# **5. Conclusion**

Since plant breeding depends on genetic variety, it is necessary to introduce innovative traits into breeding programs. The evaluated mutant populations displayed variation in morpho-physiological and agricultural characteristics. It was revealed that six mutant populations exhibited noticeably longer leaves, with LD-200-1-1-2-6 being the only mutant population to have noticeably more grain weight/hill than the parent. Six mutant populations LD-200-1-3-3-3, LD-200-1-3-3-4, LD-200-1-3-3-5, LD-200-1-3-2-1, LD-200-1-1-2-1 and LD-200-1-1-2-6 produced significantly higher grain yield hill<sup>-1</sup> which also had significantly longer panicle lengths, internode distances and heavier 1000-grain weight than the parent and also had higher levels of total chlorophyll and chlorophyll "a" in their flag leaves. In anatomical study of roots, it was found that all the 21 cross sections showed damaged cortex with different degrees except LD-200-1- 3-3-7 and LD-200-1-1-2-5. Cross sections of these two mutant populations showed no cortex damage. Even the parent grown in Chamber-1 showed minimum cortex damage. Grain yield of these two mutant populations did not differ significantly with the parent although chlorophyll "a" and total chlorophyll contents were much higher. On the basis of morpho-physiological and agronomic traits, variability was observed among the studied mutant populations. Using these divergent features in breeding programs can aid in creating breeding lines with potential morpho-physiological and agronomic qualities.

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# **Conflict of Interests**

The authors declare that there is no conflict of interests regarding the publication of this paper.

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