

Effect of Drought on Physiological Traits at Reproductive Stage in Six Rice Genotypes (*Oryza sativa* L.)

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ABSTRACT

Background and Objective: Photosynthesis, the most fundamental and intricate physiological process in all green plants, whose rate depends upon the amount of chlorophyll, is also severely affected in its reproductive phases by drought stress. Therefore, a pot experiment was performed to study drought tolerance mechanism(s) based on physiological traits in six rice genotypes.

Materials and Methods: Twelve treatments (6 genotypes×2 irrigations) were arranged in Complete Randomized Design (CRD) and the experiment was carried out at Bangladesh Institute of Nuclear Agriculture, Bangladesh Agricultural University, Mymensingh. Drought was imposed at the reproductive stage, where physiological data such as chlorophyll content and photosynthetic rate were recorded.

Results: In season 1 and 2, genotypes NERICA mutant (0.99 and 1.62) and Binadhan-13 (6.81 and 2.72) had a lesser relative reduction in chlorophyll content than in the rest of the genotypes. The relative reduction in photosynthetic rate under drought at the panicle initiation stage was lower in genotypes Binadhan-13 (3.62 and 1.87%) than in other genotypes in both seasons. Relative reduction of Photosynthetic rate at flowering under drought as compared to control was lower in genotypes NERICA mutant (3.92 and 7.68%) and Binadhan-13 (13.62 and 13.39%) than all other genotypes in both seasons.

Conclusion: Compared to control, relative reduction at 40% FC in the above parameters, Binadhan-13 and NERICA mutant genotypes were classified into drought.

KEYWORDS

Panicle initiation, flowering, chlorophyll, droughtstress, physiology, photosynthesis, rice (*Oryza sativa* L.)

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INTRODUCTION

Rice (*Oryza sativa* L.) is a major and staple food crop in many parts of the world. More than three billion people are habituated to rice feeding and rice provides 50-80% of their daily calorie intake¹. It is a drought-susceptible crop exhibiting serious deleterious effects when exposed to drought stress at critical growth stages, especially at the reproductive stage². Although plant growth is controlled by a multitude of physiological, biochemical and molecular processes, photosynthesis is a key phenomenon, which contributes substantially to plant growth and development.



The chemical energy expended in several metabolic processes is derived from the process of photosynthesis, which is capable of converting light energy into a usable chemical form of energy^{3,4}. Drought stress seriously affects plant growth and development⁵.

And it results in various physiological changes including reduced PAR, photosynthetic rate and pigmentation, resulting in decreased Water Use Efficiency (WUE) and growth before plant senescence⁶⁻⁸. These physiological parameters and yield components could be used as criteria for improving drought stress in different crops⁹.

Hence, the present study was conducted to study the impacts of drought stress at the reproductive stage on the photosynthetic rate and chlorophyll content of six rice genotypes.

MATERIALS AND METHODS

Study area: The experiment was carried out at the Crop Physiology Laboratory and Field Laboratory of the Division of the Crop Physiology, Bangladesh Institute of Nuclear Agriculture and Crop Botany Laboratory and Field Laboratory, Department of Crop Botany, Faculty of Agriculture, Bangladesh Agricultural University, Mymensingh from 2013-2015.

Materials

Plant materials: The pot experiment was conducted with six rice genotypes viz. Binadhan-13, BRRI dhan34, Ukunimadhu, RM-100-16, Kalizira and NERICA mutant collected from different organizations in the country such as Bangladesh Institute of Nuclear Agriculture (BINA), Bangladesh Rice Research Institute (BRRI), Bangladesh Agricultural Research Institute (BARI) and Bangladesh Agricultural Development Corporation (BADC).

Methods

Experimental treatments: The experiment was set in a two-factorial CRD with three replications in two seasons. The first factor was rice genotypes and the second factor was irrigations: control (100% Field Capacity) and drought (40% FC) stresses treatments. Drought (40% FC) was imposed 50 days after transplanting when plants had attained the panicle initiation stage as naturally drought starts at this stage in our climate and this stage can be treated as the initiation stage of the reproductive stage and continued till maturity.

Determination of field capacity and drought stress: The soil moisture stresses were calculated in the form of field capacity percentage. The Field Capacity (FC) of the soils used in the pots was determined by the Gravimetric Method¹⁰ FC of the soil was used as 100 and 40% of FC as drought stress.

Maintenance of different field capacity levels: Before starting stress imposition, all the pots were maintained at field capacity. Drought stress was imposed by maintaining pot soil water at 40% Field Capacity (FC) i.e., 40% field capacity was at the panicle initiation stage for seven days and discontinued after the stress was over^{11,12}. Control treatment at 100% FC was maintained from transplanting to maturity.

Drought imposition and plant establishment: Water stress application was started 50 days after transplanting when plants were attained at the panicle initiation stage, a certain amount of water according to the treatments was applied in each pot with the help of a weighing scale and continued for 7 days.

Drought estimation by gravimetric method of the experimental soil

100 g fresh soil contains 24 g water i.e., 76 g oven dry soil

For 1300 g fresh soil, actual soil was = $76 \times 13000 / 100 = 9 \text{ kg } 880 \text{ g}$

So, 13 kg of fresh soil contains 9 kg 880 g of actual soil

For 100% FC

76 g of soil contain 24 g of water

9880 g soil contain = $24 \times 9880 / 76 = 3120 \text{ gm} = 3 \text{ kg } 120 \text{ g}$

For 40% FC

Water required, $3 \text{ kg } 120 \text{ g} \times 0.40 = 1248 \text{ gm} = 1 \text{ kg } 248 \text{ g}$

For 100% FC

Every pot required = Actual soil + Water required for 100% FC + Pot wt + Plant wt (Growing plant) = 9 kg 880 g + 3 kg 120 g + 500 g + 10 g = 13 kg 510 g

For 100% FC, pot wt 13 kg 510 g

For 40% FC, pot wt = $(13 \text{ kg } 510 \text{ g} \times 0.40) = 11 \text{ kg } 700 \text{ g}$

Determination of photosynthetic rate: Rates of photosynthesis were measured after 7 days of drought imposing using a Portable Photosynthetic system (Model: Li-6400XT, I-COR Biosciences, Lincoln, NE 68504, USA) of the 3rd leaf (one leaf per plant) at the reproductive stage.

Measurements of chlorophyll content: Chlorophyll content of flag leaves was measured after 7 days of drought imposing using the SPAD meter (Model: SPAD 502, 2-16 FA YUEN, Kowloon, HK) of 3rd leaf (one leaf per plant) at the reproductive stage.

Statistical analysis: The percent reduction of each parameter under drought stress compared to control was calculated for each genotype using the formula:

$$\text{Relative increase or decrease (\%)} = \frac{\text{Data for control treatment} - \text{Data for drought treatment}}{\text{Data for control treatment}} \times 100$$

Mathematically, (-)ve value indicates an increase and (+)ve one decrease. The collected data were analyzed statistically following the Completely Randomized Design by an R software programme developed by [10]. The treatment means were adjudged by Duncan's Multiple Range Test (DMRT).

RESULTS

The combined effect of drought and genotype on chlorophyll content and the photosynthetic rate at the reproductive stage under drought stress were significant ($p \leq 0.05$) (Table 1 and 2). The relative reduction in percentage (figures in the parenthesis) of this variable under drought as compared to control is shown (Table 1 and 2). Chlorophyll content under drought at the panicle initiation stage was lower in genotypes NERICA mutant (0.99), Binadhan-13 (6.81) and Kalizira (1.70) than rest of the genotypes (RM 100-16-8.74, Ukunimodhu-7.02, BRRI dhan-34-8.46) in season-1. In season 2, on the other hand, genotypes NERICA mutant (1.62) and Binadhan-13 (2.72) had lesser relative reduction than the rest genotypes (RM 100-16-4.66, Ukunimodhu-4.98, BRRI dhan-34-8.01 (Table 1). These results indicated that chlorophyll content varied between the seasons and genotypes and importantly genotypes NERICA mutant and Binadhan-13 had a significantly lower decrease in chlorophyll content in season 1 and season 2, respectively. The relative reduction in photosynthetic rate under drought at the panicle initiation stage was lower in genotypes Binadhan-13 (3.62) and Ukunimodhu (3.82) than in the rest genotypes (Kalizira-15.93, RM-100-16- 9.14, BRRI dhaan- 14.36 and NERICA Mutant-15.16) in season 1. In season 2, only genotype Binadhan-13 had a lower relative reduction (1.87%) than all other genotypes (Table 1).

Table 1: Combined effect of genotype and drought with control irrigations on physiological traits in six rice genotypes at the panicle initiation stage

Genotype	Irrigation	Chlorophyll content (SPAD value)	Photosynthetic rate (Pn) ($\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$)
2013			
Binadhan-13	Control (100% FC)	42.70 ^{a+}	38.70 ^{ab}
	Drought (40% FC)	39.70 ^a (6.81) ⁺⁺	37.30 ^{a-c} (3.62)
Kalizira	Control (100% FC)	41.13 ^a	39.93 ^a
	Drought (40% FC)	40.43 ^a (1.70)	33.57 ^{a-d} (15.93)
RM 100-16	Control (100% FC)	42.60 ^a	35.33 ^{a-d}
	Drought (40% FC)	38.97 ^a (8.74)	32.10 ^{b-e} (9.14)
Ukunimodhu	Control (100% FC)	40.73 ^a	29.03 ^{cde}
	Drought (40% FC)	37.87 ^a (7.02)	28.73 ^{de} (3.82)
BRRI dhan-34	Control (100% FC)	42.57 ^a	33.20 ^{a-d}
	Drought (40% FC)	38.97 ^a (8.46)	28.43 ^{de} (14.36)
NERICA mutant	Control (100% FC)	40.50 ^a	29.87 ^{de}
	Drought (40% FC)	40.10 ^a (0.99)	24.63 ^e (15.16)
2014			
Binadhan-13	Control (100% FC)	38.57 ^a	38.03 ^a
	Drought (40% FC)	37.52 ^{b-d} (2.72)	37.32 ^a (1.87)
Kalizira	Control (100% FC)	37.59 ^{ab}	35.47 ^{ab}
	Drought (40% FC)	34.12 ^d (9.23)	33.94 ^b (4.31)
RM 100-16	Control (100% FC)	37.11 ^{abc}	36.47 ^{ab}
	Drought (40% FC)	35.38 ^{b-d} (4.66)	33.86 ^b (7.25)
Ukunimodhu	Control (100% FC)	38.34 ^a	35.68 ^{ab}
	Drought (40% FC)	36.43 ^{a-d} (4.98)	33.14 ^b (7.11)
BRRI dhan-34	Control (100% FC)	37.93 ^{ab}	36.49 ^{ab}
	Drought (40% FC)	34.89 ^{cd} (8.01)	34.51 ^b (5.42)
NERICA mutant	Control (100% FC)	38.19 ^a	38.03 ^a
	Drought (40% FC)	37.57 ^{ab} (1.62)	35.42 ^{ab} (6.86)

+ : Data were separately analyzed for the years 2013 and 2014, in a year in each column, figures having a common letter(s) do not differ significantly at $p \leq 0.05$ as per DMRT and ++ : Figures within parenthesis indicate a % decrease at 40% FC compared to control

Table 2: Combined effect of genotype and drought with control irrigations on physiological traits in six rice genotypes at the flowering stage

Genotype	Irrigation	Chlorophyll (SPAD value)	Photosynthetic rate (Pn) ($\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$)
2013			
Binadhan-13	Control (100% FC)	40.01 ^{a+}	23.27 ^c
	Drought (40% FC)	39.00 ^{ab} (2.52) ⁺⁺	20.10 ^d (13.62)
Kalizira	Control (100% FC)	37.77 ^{abc}	20.23 ^d
	Drought (40% FC)	37.41 ^{abc} (0.95)	13.85 ^f (31.53)
RM 100-16	Control (100% FC)	37.98 ^{abc}	20.13 ^d
	Drought (40% FC)	36.90 ^{bc} (2.84)	12.75 ^g (36.66)
Ukunimodhu	Control (100% FC)	36.07 ^c	20.32 ^d
	Drought (40% FC)	35.96 ^c (0.30)	13.49 ^f (33.61)
BRRI dhan-34	Control (100% FC)	38.09 ^{abc}	20.17 ^d
	Drought (40% FC)	37.21 ^{abc} (2.31)	12.77 ^g (36.69)
NERICA mutant	Control (100% FC)	37.66 ^{abc}	36.25 ^a
	Drought (40% FC)	36.63 ^{bc} (2.73)	34.83 ^b (3.92)
2014			
Binadhan-13	Control (100% FC)	42.41 ^a	24.33 ^c
	Drought (40% FC)	41.34 ^a (2.52)	21.07 ^d (13.39)
Kalizira	Control (100% FC)	40.03 ^a	21.15 ^d
	Drought (40% FC)	39.66 ^a (0.92)	14.46 ^e (31.63)
RM 100-16	Control (100% FC)	40.25 ^a	21.05 ^d
	Drought (40% FC)	39.11 ^a (2.83)	13.32 ^e (36.72)
Ukunimodhu	Control (100% FC)	38.23 ^a	21.23 ^d
	Drought (40% FC)	37.13 ^a (2.88)	14.09 ^e (33.63)
BRRI dhan-34	Control (100% FC)	40.38 ^a	21.10 ^d
	Drought (40% FC)	39.45 ^a (2.30)	13.35 ^e (36.73)
NERICA mutant	Control (100% FC)	40.75 ^a	41.28 ^a
	Drought (40% FC)	40.43 ^a (0.79)	38.11 ^b (7.68)

+ : Data were separately analyzed for the years 2013 and 2014, in a year in each column, figures having a common letter(s) do not differ significantly at $p \leq 0.05$ as per DMRT and ++ : Figures within parenthesis indicate a % decrease at 40% FC compared to control

Results suggest that the genotype Binadhan-13 showed a lower reduction in photosynthetic rate in both years thereby conferring a greater degree of drought tolerance. Relative reduction of flag leaf chlorophyll content at flowering under drought as compared to control was lower in genotypes Kalizira (0.95) and Ukunimodhu (0.30) than all remainders (Binadhan-13- 2.52, RM 100-16-2.84, BRR1 dhan-34-2.31, NERICA Mutant- 2.73) in 2013. In 2014, genotypes NERICA mutant (0.79) and Kalizira (0.92) had lower relative reduction (average of 0.86%) than all others as Binadhan-13(2.52), RM 100-16 (2.83), Ukunimadhu (2.88) and BRR1 dhan-34 (2.30) (Table 2). Relative reduction of Photosynthetic rate (Pn) at flowering under drought as compared to control was lower in genotypes Binadhan-13 (13.62) and NERICA mutant (3.92) than all other genotypes as Kalizira (31.53), Ukunimadhu (33.61), RM 100-16 (33.66) and BRR1 dhan-34 (36.69) in 2013. In 2014, once again genotypes Binadhan-13 (13.39) and NERICA mutant (7.68) had a lower reduction than all other genotypes as Kalizira (31.63), RM 100-16 (36.72), Ukunimadhu (33.63) and BRR1 dhan-34 (36.73) (Table 2).

The results of Pn indicated that this trait had consistently discriminated the two genotypes, Binadhan-13 and NERICA mutant, being drought tolerant.

DISCUSSION

In season 1 and 2, genotypes NERICA mutant and Binadhan-13 had a lesser relative reduction in chlorophyll content and photosynthetic rate than in the rest of the genotypes at the panicle initiation stage. Relative reduction of photosynthetic rate and chlorophyll content at flowering under drought as compared to control was lower in genotypes NERICA mutant and Binadhan-13 than all other genotypes in both seasons. A differential decrease in the physiological response of upland rice varieties was also observed¹³⁻¹⁵. The decrease in chlorophyll content is a commonly observed phenomenon under drought stress¹⁶. In this study, Binadhan-13 and NERICA mutant genotypes had shown increased levels of chlorophyll and such enhanced accumulation of chlorophyll under drought stress was also observed in cereals by others^{17,18}. Like chlorophyll reduction under drought photosynthetic rate was also significantly reduced. Drought stress reduces the photosynthetic rate by stress-induced stomatal or nonstomatal limitations^{19,20}. In summary, drought stress not only limits chlorophyll content but also photosynthetic rate. Based on the lower relative reduction of all the above physiological variables, Binadhan-13 and NERICA mutant genotypes demonstrated better traits compared to other genotypes as they become tolerant to drought. Drought stress decreases the photosynthetic rate and disrupts the carbohydrate metabolism and sucrose level in leaves.

This is presumably due to drought stress-induced increased activity of acid invertase²¹ stating that limited photosynthesis in the flag leaves may affect reproductive development. Present findings have also shown photosynthesis rate and chlorophyll content decreased at the reproductive stage under drought. Thus, in drought stress conditions both photosynthesis rate and chlorophyll content were also decreased at the reproductive stage. These may be the most popular parameters used to identify drought tolerance in rice breeding programs. The findings are useful in harmony with different agronomic parameters during the screening of rice varieties or while developing new rice varieties for drought-prone areas.

CONCLUSION

Two genotypes (BINADhan-13 and NERICA) showed lower reduction under drought compared to control in physiological parameters compared to all remainders. Finally, key physiological features like photosynthesis and chlorophyll content were investigated, which showed significant tolerant genotypes and all of these key physiological features interactions. Moreover, their relative reduction under drought as compared to respective control showed lower in general in Binadhan-13 and NERICA mutant genotypes. This suggests their tolerance level to drought in key physiological features.

SIGNIFICANCE STATEMENT

This study discovers biochemical traits like increased proline and total sugar contents which may be considered an important index of drought-tolerant mechanisms in aromatic rice genotypes. This study will help the researcher to uncover the critical areas of yield attributes of rice genotypes under drought stress particularly at the reproductive stage which can be beneficial for drought-prone areas of Bangladesh, that many researchers were not able to explore. Thus a new theory on the development of drought-tolerant rice genotypes may be arrived at.

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