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Effect of Nitrobenzene on the Growth and Yield of Rice

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ABSTRACT

A field experiment was conducted at two locations (Gazipur and Satkhira) during the Boro (dry) season in 2020 to study the performance of Nitrobenzene on the growth and yield of rice. Four different concentrations of Nitrobenzene (2.0 mL L^{-1}, 2.5 mL L^{-1}, 3.0 mL L^{-1} and 3.5 mL L^{-1}) as foliar spray with recommended chemical fertilizer (RF) were tested on BRRI dhan89 under wetland condition. Application of Nitrobenzene at different concentrations with RF showed significant effect on rice growth and yield. At both the locations, foliar application of Nitrobenzene (2 mL L^{-1}) + RF produced higher number of tiller/m^2, panicle/m^2 and grain/panicle over RF. At Gazipur site, Nitrobenzene (2 mL L^{-1}) + RF resulted in significant increase of grain and straw yield of BRRI dhan89 over the RF only whereas, at Satkhira site, no significant yield benefit with Nitrobenzene was found over RF. In terms of rice growth, yield and economic point of view, foliar application of Nitrobenzene at low concentration (2 mL L^{-1}) with recommended fertilizer showed positive effect on the growth and yield of BRRI dhan89 in this study. However, since Nitrobenzene is a new type of plant stimulant, detailed studies on its mode of action, interaction with other plant growth hormones and environmental effects are required to get an insight on rice response to Nitrobenzene and its widespread application in rice cultivation.

Key words: Rice growth, rice yield, foliar spray, economic benefit

INTRODUCTION

In Bangladesh, farmers generally use chemical fertilizers with little or no use of organic manures to meet the rice nutrient requirement, which results in degraded soil health and decreased rice yield (Sarkar *et al.*, 2016). Farmers could not use sufficient organic manures as they are required in bulk amount and are less available. Therefore, to curtail chemical fertilizer use as well as to increase rice yield an efficient and sustainable alternative is imperative. To solve this problem, plant growth regulator could be an efficient option. Plant growth regulators are widely used for their ability to increase crop growth and yield. Among the plant growth regulators, nitrobenzene found as effective in improving crop production in some studies (Aziz and Miah, 2009; Kohombange *et al.*, 2017; Kohombange *et al.*, 2019). Nitrobenzene is a combination of nitrogen and plant growth regulators, extracted from sea weeds that act as plant energizer, flowering stimulant and yield booster. Nitrobenzene is quickly absorbed into the plants and it influences the biochemical pathway of plants to uptake more nutrients from the soil. It also promotes vegetative growth and yield by increasing nutrient use efficiency, flowering and retention of flower and fruits (Deb *et al.*, 2009). The use of plant growth regulators like nitrobenzene in rice in Bangladesh is very limited. It is imperative to properly understand the effects of Nitrobenzene on rice growth and yield before its widespread use in rice

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cultivation. Therefore, this study was conducted to evaluate the performance of Nitrobenzene on the growth and yield of rice as well as to assess the profitability of Nitrobenzene application in rice cultivation.

MATERIALS AND METHODS

A field experiment was conducted at two locations: one at Bangladesh Rice Research (BRRI) farm, Gazipur under AEZ 28 and the other one at BRRI farm, Satkhira under AEZ 13 in Boro (dry) season, 2020 (Fig. 1). Table 1 presents the initial soil properties of the experimental locations. Soil pH, organic matter, total N, available P, exchangeable K, available S and available Zn were analyzed following the methods described by Tedesco *et al.*, 1995, Walkley and Black, 1934; Bremner and Mulvaney 1982, Sparks *et al.*, 1996; Black *et al.*, 1965; Fox *et al.*, 1965 and Olsen and Ellis, 1982, respectively. Fig. 2 presents the weather data (temperature and rainfall) of the experimental locations during the crop growing period. In this study, the efficacy of Nitrobenzene (Nitrobenzene 20% w/w obtained by the trade name Flora) was tested in five treatment combinations: T_1= Recommended chemical fertilizer only (RF), T_2 = RF + Nitrobenzene @ 2.0 mL L^{-1}, T_3 = RF + Nitrobenzene 2.5 mL L^{-1}, T_4 = RF + Nitrobenzene 3.0 mL L^{-1}, and T_5 = RF + Nitrobenzene 3.5 mL L^{-1}. The recommended rates of nitrogen (N), phosphorus (P), potassium (K), sulfur (S) and zinc (Zn) were 138, 21, 75, 18 and 4 kg ha^{-1} respectively, at both the locations. The treatments were assigned in randomized complete block design with three replications. The N, P, K, S and Zn nutrients were applied as urea, triple super phosphate (TSP), muriate of potash (MoP), gypsum and zinc sulfate, respectively. Full dose of TSP, MoP, gypsum and zinc sulfate were applied at the time of final land preparation. Urea was applied into three equal splits: 1/3rd after 12 days of transplanting, next 1/3rd at maximum tillering stage and the remaining 1/3rd at panicle initiation stage. Nitrobenzene of different doses were sprayed five times on the rice plant during the growing period according to the spray schedule (Table 2). At both the locations, thirty-five days old 2-3 seedlings/hill of BRRI dhan89 were transplanted with 20 cm × 20 cm spacing. Weeding, pesticide application and other necessary intercultural operations were done when required. At maturity, the crop was harvested from the centre 5 m2 area of each plot and the grain yield was adjusted to 14% moisture content. The data of plant height, tiller and panicle number per meter, filled grain number per panicle, thousand grain weight, grain and straw yields and harvest index were recorded.

Harvest index (HI) was estimated based on grain and straw yields using the following formula:

$$Harvest\ index = \frac{Grain\ yield}{Biological\ yield} \ldots\ldots\ldots\ldots (1)$$

where, Biological yield = grain yield + straw yield.

The grain and straw samples were analyzed for N concentration by micro-Kjeldahl method (Nelson and Sommers, 1973) and P and K concentrations were anlyzed by nitric-perchloric acid digestion method (Yamakawa, 1992) to calculate the total N, P and K uptake by the rice crop. The nutrient uptake was calculated as:

$$Nutrient\ uptake\ (kg/ha) = \frac{Nutrient\ concentration\ (\%) \times Yield\ (kg/ha)}{100} \ldots\ldots\ldots\ldots (2)$$

The data related to rice crop growth and yield were subjected to analysis of variance (ANOVA) to determine the treatment effects. Least Significance Difference (LSD) at the 5% level of probability was used to separate the means for treatments effect. All the analyses were performed using Statistical Tool for Agricultural Research (STAR 2.0.1, International Rice Research Institute, Manila, the Philippines).

Economic analysis was done to estimate added return, net return and marginal benefit cost ratio (MBCR) according to the following equations:

$$Added\ return = Gross\ return_{Flora\ treated\ plot} - Gross\ return_{Flora\ control\ plot} \quad \ldots\ldots\ldots (3)$$

$$Net\ return = Gross\ return - Total\ variable\ cost \quad \ldots\ldots\ldots\ldots\ldots (4)$$

$$MBCR = \frac{Gross\ return_{Flora\ treated\ plot} - Gross\ return_{Flora\ control\ plot}}{Total\ variable\ cost_{Flora\ treated\ plot} - Total\ variable\ cost_{Flora\ control\ plot}} \quad \ldots\ldots\ldots\ldots (5)$$

Fig. 1. Study locations.

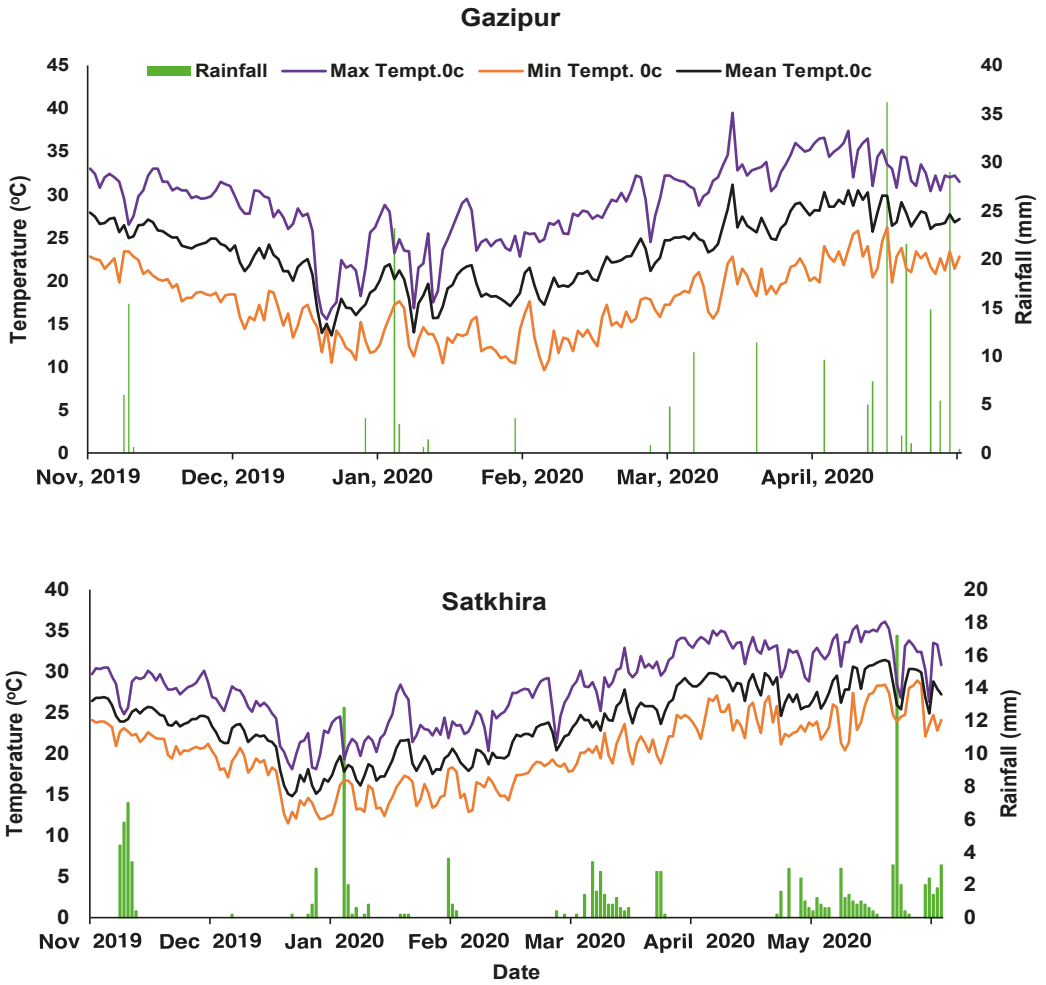

Fig. 2. Temperature and rainfall data of the study locations during crop growing period.

Table 1. Initial soil characteristics of the study locations.

Parameter	BRRI farm, Gazipur	BRRI farm, Satkhira
pH (1 : 2.5)	6.80	7.30
Organic C (%)	1.20	1.40
Total N (%)	0.11	0.13
Available P (mg/kg)	8.20	11.00
Exchangeable K (cmol/kg)	0.17	0.20
Available S (mg/kg)	19.00	30.00
Available Zn (mg/kg)	3.70	1.10

Table 2. The spray schedule of Nitrobenzene.

Spray schedule	Spray time
First spray	7 days before transplanting
Second spray	25 days after transplanting
Third spray	50 days after transplanting
Fourth spray	75 days after transplanting
Fifth spray	100 days after transplanting

RESULTS AND DISCUSSION

Effect of foliar application of Nitrobenzene on growth parameters

Foliar application of Nitrobenzene at different rates showed no significant effect on plant height of BRRI dhan89 at Gazipur and Satkhira sites. However, at both the locations the tiller number m^{-2} varied significantly with Nitrobenzene spray of varying rates. At both the locations, T_2 (RF + Nitrobenzene @ 2 mL L^{-1}) produced the highest tiller number m^{-2} compared to other treatments. The T_2 treatment increased tiller number m^{-2} by 11% and 10% over the recommended chemical fertilizer (T_1) at Gazipur and Satkhira sites, respectively (Table 3). In this study, foliar application of Nitrobenzene in combination with chemical fertilizer showed positive effect on tiller number of BRRI dhan89 at the study locations at low concentration (2 mL L^{-1}). The enhanced plant growth with Nitrobenzene may be attributed to synergistic relationship between Nitrobenzene and other plant hormones namely gibberellins, auxin and cytokinin (Kohombange *et al.*, 2019). Gibberellins and auxin influence stem elongation while auxin and cytokinin influence lateral growth of parenchyma cells in stem so that the plant girth increases (Nickell, 1982). Moreover, Nitrobenzene promotes plant nutrient uptake from the soil and nutrient use efficiency by influencing the biochemical pathway of the plant resulting in improved plant growth (Deb *et al.*, 2012). However, in our study at both locations the total uptake of N, P and K showed no significant variation with the treatments applied.

Table 3. Effects of foliar application of Nitrobenzene on growth and yield parameter of BRRI dhan89.

Treatment*	Plant height (cm)		Tiller m^{-2}	
	Gazipur	Satkhira	Gazipur	Satkhira
T_1	102.67	110.44	301 b	288 bc
T_2	102.67	110.00	335 a	318 a
T_3	103.00	112.56	284 b	301 b
T_4	101.00	110.44	314 ab	298 bc
T_5	101.00	107.33	304 b	284 c
$LSD_{0.05}$	2.00	4.00	31	16
CV (%)	1.03	2.00	*5.31*	*3.83*

*T_1= Recommended chemical fertilizer (RF), T_2= RF + Nitrobenzene (2.0 mL L^{-1}), T_3= RF + Nitrobenzene (2.5 mL L^{-1}), T_4 = RF + Nitrobenzene (3.0 mL L^{-1}), T_5 = RF + Nitrobenzene (3.5 mL L^{-1}).

Effect of foliar application of Nitrobenzene on yield parameters

Table 4 presents the effect of different treatments on yield components of BRRI dhan89. Application of Nitrobenzene at different rates in combination with recommended fertilizer significantly influenced the panicle number m^{-2}. At Gazipur site, the highest panicle number m^{-2} (329) were recorded with T_2 (RF + Nitrobenzene @ 2 mL L^{-1}), which was statistically similar to T_4 (RF + Nitrobenzene @ 3 mL L^{-1}). At Satkhira site, the T_2 treatment also produced the highest number (308) of panicles m^{-2} compared to the other treatments. The T_2 treatment increased panicle number m^{-2} by 12% and 9% over the recommended fertilizer only (T_1) at Gazipur and Satkhira sites, respectively. The number of grains per panicle and sterility (%) differed significantly with varying levels of Nitrobenzene spray at both the locations. At Gazipur site, the highest number (147) of filled grain per panicle was found with T_2 treatment which was statistically similar with T_3 and T_4 treatments. At Satkhira site, the highest number (133) of grain per panicle was found with T_2 than the other treatments. At both the locations the grain sterility (%) significantly reduced with the T_2 treatment and was higher with T_5. The thousand grain weight of BRRI dhan89 did not differ with the applied treatments. Number of panicle m^{-2} and filled grain were significantly higher with Nitrobenzene sprayed at 2 mL L^{-1} compared to only chemical fertilizer, which indicates that Nitrobenzene might have positive effect on rice flowering and grain filling. The application of Nitrobenzene and its immediate transport to the auxiliary buds would have resulted in a better sink for the quick mobilization of photo-assimilates. The increased number of filled grain might be influenced by triggering of such metabolic processes and the increased accumulation of carbohydrates into the rice grain which resulted in reduced grain sterility. The sterility of BRRI dhan89 was comparatively higher in Satkhira than that of Gazipur, which might be due to the variation in climatic condition between the two locations.

Table 4. Effects of foliar application of Nitrobenzene on yield parameters of BRRI dhan89.

Treatment*	Panicle m^{-2}		Grain/panicle		1000 grain wt.		Sterility %	
	Gazipur	Satkhira	Gazipur	Satkhira	Gazipur	Satkhira	Gazipur	Satkhira
T_1	294	282	134	116	23.34	24.02	11	20
T_2	329	308	147	133	23.04	23.52	10	18
T_3	278	288	138	117	23.01	23.31	10	18
T_4	304	288	125	113	22.90	23.65	13	21
T_5	296	274	114	112	23.27	24.12	15	31
$LSD_{0.05}$	*31*	*15*	*17*	*17*	*1.10*	*1.20*	*4*	*4*
CV (%)	*5.53*	*3.63*	*6.94*	*7.40*	*2.78*	*2.45*	*19*	*10*

*T_1= Recommended chemical fertilizer (RF), T_2= RF + Nitrobenzene (2.0 mL L^{-1}), T_3= RF + Nitrobenzene (2.5 mL L^{-1}), T_4 = RF + Nitrobenzene (3.0 mL L^{-1}), T_5 = RF + Nitrobenzene (3.5 mL L^{-1}).

Effect of foliar application of Nitrobenzene on grain and straw yield

The grain and straw yields of BRRI dhan89 significantly differed with the different treatments and table 5 presents the results. At Gazipur site, the T_2 (RF + Nitrobenzene @ 2 mL L^{-1}) treatment produced the highest grain and straw yields than the other treatments. The T_2 treatment increased grain yield by about 11% over recommended fertilizer only (T_1). At Satkhira site, the highest grain yield was found with T_2 (RF+ Nitrobenzene @ 2 mL L^{-1}) which was statistically identical to T_1, T_3 and T_4 treatments. The T_2 treatment increased grain yield by about 5% over T_1. The highest straw yield was found with T_3 (RF+ Nitrobenzene @ 2.5 mL L^{-1}) which was statistically similar to T_1, T_2 and T_5 treatments. Irrespective of location the harvest index of BRRI dhan89 varied between 0.50-0.53 and there was significant variation among the different treatments. The increased grain yield of BRRI dhan89 with Nitrobenzene sprayed at a concentration of 2 mL L^{-1} was attributed to the higher number of tiller, panicle and filled grain with this treatment. Deb *et al.*, 2012 and Kohombange *et al.*, 2019 reported that Nitrobenzene increased flowering and fruit sets in tomato and cucumber, respectively which support our findings. However, the significant yield increase was observed only at Gazipur site indicating that effect of Nitrobenzene may vary with locations because of variation in initial soil fertility (Table 1). Our study clearly indicate that application of flora at concentration above 2 mL L^{-1} showed no significant effect on rice plant growth and yield. Guo *et al.*, 2010 found that in solution culture the growth of soybean seedlings increased at low concentration of 5 mg L^{-1} and after 10 mg L^{-1} showed genotoxic effects. Moreover, higher concentrations of nitrobenzene in water bodies are hazardous to aquatic organism and plants by irrigation (Guo *et al.*, 2010).

Table 5. Effects of foliar application of Nitrobenzene on grain and straw yield (t/ha) of BRRI dhan89.

Treatment*	Grain yield (t ha^{-1})		Straw yield (t ha^{-1})		Harvest index		% Grain yield increased over recommended chemical fertilizer	
	Gazipur	Satkhira	Gazipur	Satkhira	Gazipur	Satkhira	Gazipur	Satkhira
T_1	6.79 b	6.67ab	6.19 b	6.54ab	0.52	0.50		
T_2	7.53 a	6.99 a	6.85 a	6.86 a	0.52	0.50	10.82	4.66
T_3	6.93 b	6.84 ab	6.33 b	6.87 a	0.52	0.50	2.01	2.44
T_4	6.82 b	6.54 ab	6.25 b	6.24 b	0.52	0.51	0.46	-2.04
T_5	6.85 b	6.44 b	6.17 b	6.52 ab	0.53	0.50	0.91	-3.52
$LSD_{0.05}$	*0.43*	*0.28*	*0.46*	*0.49*				
CV (%)	*4.27*	*4.69*	*3.35*	*4.95*				

*T_1= Recommended chemical fertilizer (RF), T_2= RF + Nitrobenzene (2.0 mL L^{-1}), T_3= RF + Nitrobenzene (2.5 mL L^{-1}), T_4 = RF + Nitrobenzene (3.0 mL L^{-1}), T_5 = RF + Nitrobenzene (3.5 mL L^{-1}).

Effects of foliar application of Nitrobenzene on nutrient uptake of BRRI dhan89

Application of Nitrobenzene at different concentrations showed no significant effect on the total uptake of nitrogen (N), phosphorus (P) and potassium (K) by BRRI dhan89 at both the locations. At Gazipur site, the total uptake of N, P and K ranged between 108.63 – 126.70 kg ha^{-1}, 20.69 – 23.80 kg ha^{-1} and 99.47 – 113.73 kg ha^{-1}, respectively. At Satkhira site, the total uptake of N, P and K ranged between 109.78 – 120.72 kg ha^{-1}, 17.77 – 21.09 kg ha^{-1} and 84.70 – 95.11 kg ha^{-1}. respectively.

Economic analysis

Tables 6 and 7 present the calculated total variable cost (TVC), gross return, added return, net return and marginal benefit cost ratio (MBCR) of BRRI dhan89 with different treatments. The total variable cost was calculated considering the cost for fertilizer, fertilizer application, Nitrobenzene spray and labour for the additional products (Table 6). At both the locations, foliar application of Nitrobenzene (2mL L^{-1}) with the recommended fertilizer resulted in the highest gross return, added return and net return. Moreover, application of Nitrobenzene at higher rates was not profitable as the added and net returns were much lower with higher rates of Nitrobenzene. At both the locations, similar results were also found for MBCR (Table 7).

Table 6. Economic analysis of foliar application of Nitrobenzene on BRRI dhan89 at two different locations of Bangladesh.

Total variable cost (TVC) in Tk/ha

Treatment*	Fertilizer cost		Fertilizer application cost		Nitrobenzene spray cost		Labor cost for additional products		Total variable cost (BDT ha^{-1})	
	Gazipur	Satkhira	Gazipur	Satkhira	Gazipur	Satkhira	Gazipur	Satkhira	Gazipur	Satkhira
T_1	14560	14560	800	800	0	0	0	0	15360	15360
T_2	16180	16180	800	800	2000	2000	5580	2534	24560	21514
T3	16585	16585	800	800	2000	2000	1134	1975	20519	21359.8
T4	16990	16990	800	800	2000	2000	359.6	0	20149.6	19790
T5	17395	17395	800	800	2000	2000	184.4	0	20379.4	20195

*T_1= Recommended chemical fertilizer (RF), T_2= RF + Nitrobenzene @ 2.0 mL L^{-1}, T_3= RF + Nitrobenzene @ 2.5 mL L^{-1}, T_4 = RF + Nitrobenzene@ 3.0 mL L^{-1}, T_5 = RF + Nitrobenzene @ 3.5 mL L^{-1}. Fertilizer cost included price of chemical fertilizer and Nitrobenzene. Urea = Tk 16/kg, TSP = Tk 22/kg, MoP= Tk15/kg, Gypsum= Tk 25/kg, Zinc= Tk 200/kg, Nitrobenzene (Flora) = Tk 810/lit. Labor wage= Tk 400/day. Two additional man days/ha are required for applying fertilizer, ten-man days/ha for per ton additional products and five-man days/ha for spraying Nitrobenzene.

Table 7. Gross and net return in Tk/ha and marginal benefit cost ratio (MBCR)

Treatment*	Yield				Total variable cost		Return						MBCR	
	Grain		Straw				Gross		Added		Net			
	Gazipur	Satkhira	Gazipur	Satkhira	Gazipur	Satkhira	Gazipur	Satkhira	Gazipur	Satkhira	Gazipur	Satkhira	Gazipur	Satkhira
T_1	6.79	6.67	6.19	6.54	15360	15360	188989	186631	-	-	173629	171271	-	-
T_2	7.53	6.99	6.85	6.86	24560	21514	209421	195359	20432	8729	184861	173845	2.22	1.42
T_3	6.93	6.84	6.33	6.87	20519	21360	192830	191535	3841	4904	172311	170175	0.74	0.82
T_4	6.82	6.54	6.25	6.24	20150	19790	189918	182484	929	-4147	169768	162694	0.19	-0.94
T_5	6.85	6.44	6.17	6.52	20379	20195	190557	180494	1568	-6137	170178	160299	0.31	-1.27

*T_1 = Recommended Fertilizer (RF), T_2 = RF + Nitrobenzene @ 2.0 mL L^{-1}, T_3 = RF + Nitrobenzene @ 2.5 mL L^{-1}, T_4 = RF + Nitrobenzene @ 3.0 mL L^{-1}, T_5 = RF + Nitrobenzene @ 3.5 mL L^{-1}; GS = Gazipur site, SS = Satkhira Site. Rice grain price = Tk 26/kg, Rice straw price = Tk 2/kg.

CONCLUSION

Foliar application of Nitrobenzene significantly affected the growth and yield of BRRI dhan89. Among different concentrations of Nitrobenzene tested, Nitrobenzene sprayed at a concentration of 2 mL L^{-1} with recommended chemical fertilizer (RF) resulted in significantly higher tiller and panicle number and grain number. Compared to only RF, combined application of nitrobenzene (2mL L^{-1}) with RF produced significant yield benefit of BRRI dhan89 only at Gazipur site. Irrespective of location, application of Nitrobenzene at higher rates showed no significant advantages on the growth and yield of BRRI dhan89. In this study, foliar spray of Nitrobenzene with a rate of 2mL L^{-1} performed better in terms of plant growth and yield of BRRI dhan89. However, this study was a single season experiment and conducted only at two locations, thus multilocational trials with long term study which is reqiured to draw a concrete conclusion on rice response to Nitrobenzene.

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