

**EFFECTS OF TIBA AND RHIZOBIUM APPLICATION ON GROWTH,
YIELD AND BIOCHEMICAL COMPONENTS OF BRRI DHAN-55
(*ORYZA SATIVA* L.)**

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Abstract

Effects of TIBA (10, 25, 50 ppm) and *Rhizobium* inoculation on growth, yield and biochemical components of BRRI Dhan-55 were investigated. Results showed that application of 10 ppm TIBA produced tallest plant, higher number of tillers and leaves per plant although statistically identical to control. The *Rhizobium* and TIBA treatments had mostly retarding effects on dry weight of leaves, shoots and roots where the lowest values was obtained from *Rhizobium* application. Leaf area ratio was positively affected by TIBA and *Rhizobium* treatments except due to 10 ppm where, significantly maximum value was noted from *Rhizobium* treatment. Specific leaf weight, relative growth rate and net assimilation rate were negatively responded following all treatments and the least value was also recorded from *Rhizobium* treatment in each cases. Yield attributes and yield of BRRI Dhan-55 were both positively and negatively influenced by *Rhizobium* and TIBA treatments. The highest harvest index was found in *Rhizobium* treated plants. The only increase in yield per plant due to 10 ppm TIBA was 2.48 % over the control but statistically similar to control. Results showed that *Rhizobium* inoculation had showed lowest value in protein content of leaves and seeds. Application of TIBA treatments had significant positive effects on protein content of leaves at flowering stage. However, the influence was rather negative at both tillering and grain filling stages. The 25 ppm TIBA resulted significantly maximum protein content of seeds followed by 10 ppm.

Introduction

Rice (*Oryza sativa* L.) covers about 77% of agricultural land and engages about 70% of total population of Bangladesh and the contribution of rice to GDP is about 18% (Chowdhury and Hassan 2013). It is the single most important crop which is a primary source for half of the world's population. Phytohormones are the magic chemicals that could increase agricultural production at an unprecedented rate and help in removing and circumventing many of the barriers imposed by genetics and environment. Investigations throughout the world revealed that the application of TIBA (2,3,5 tri-iodobenzoic acid) at optimum concentration had stimulatory effects on growth, yield and biochemical components of different plants (Adam and Jahan 2014, Jahan and Khan 2014, Munira 2015, Islam and Jahan 2019). But, reports regarding the effect of TIBA on rice crops are limited from both home and abroad (Adam *et al.* 2015, Adam *et al.* 2018).

On the other hand, Dobbelaere *et al.* (2003), Alikhani *et al.* (2006) reported various mechanisms in plant growth promoting activities of *Rhizobium* sp. Findings of different investigation revealed that plant growth promoting bacteria have potential enhancement effect on growth and yield of some rice varieties (Hussain *et al.* 2009, Tan *et al.* 2015). The *Rhizobium* sp. can also help in increasing the production of non-legume plants by promoting growth and enduring the abiotic and biotic stress (Oladele and Awodun 2014).

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In Bangladesh, very few experiments have been done with rice using *Rhizobium* inoculation (Biswas *et al.* 2000, Hussain *et al.* 2009). Therefore, an attempt was taken to evaluate the responses of TIBA and *Rhizobium* inoculation on the growth, yield and biochemical components of rice, var. BRRI Dhan-55.

Materials and Methods

A pot experiment was conducted at the botanical garden of the Department of Botany, Jagannath University, Dhaka. A high yielding transplanting popular Aush variety, BRRI Dhan-55 which was released in 2011 was used in this investigation. The experiment was laid out in RCBD with five replications for each treatment. Seeds of BRRI Dhan-55 were collected from Bangladesh Rice Research Institute (BRRI), Gazipur whereas, *Rhizobium* strain (*Rhizobium* BARI Rca 259) was collected. The experiment consisted of five treatments *viz.* control (without *Rhizobium sp.* and TIBA, seed inoculated with *Rhizobium sp.*, foliar application of 10, 25 and 50 ppm TIBA. Seeds were surface sterilized with Captan 200 before sowing to avoid fungal infection and were kept in darkness for 48 hrs for germination. In case of *Rhizobium* inoculation each seed was inoculated directly with 1 ml of bacterial culture (10^8 CFU/ml) grown in nutrient broth (Aarab *et al.* 2015). After inoculation the inoculated seeds were kept for 2 hrs (Hussain *et al.* 2009). Seeds were sown in the seed bed and seedlings were transplanted to experimental pots (25×30 cm) at 4-leaf stage at the age 25 days after sowing. Each pot was filled with 9.0 kg air dried soil. Three hills were placed in each pot, each hill containing one seedling. Cultural practices *viz.* thinning, irrigation and weeding and fertilizer application were done following Hand Book of Agricultural Technology (Chowdhury and Hassan 2013) and Fertilizer Recommendation Guide (2012) respectively. Cow dung was also mixed with soil uniformly during pot preparation. Foliar application of TIBA was done in sunny early morning at 30 days after transplanting. Growth parameters *viz.* plant height, number of tillers and leaves per plant, dry weight of leaves, shoot and root per plant, leaf area ratio, specific leaf weight, relative growth rate and net assimilation rate and all yield parameters were recorded at harvest. Whereas, Leaf area ratio, specific leaf weight, relative growth rate and net assimilation were calculated using classical growth analysis methods (Radford 1967, Gardner *et al.* 1985). Pigment and protein contents of fresh leaves were determined at three different stages *viz.* tillering, flowering and grain filling stages following standard procedures (Mckinney 1940, Lowry *et al.* 1951, von Wettstein 1957, Maclachalan and Zalik 1963). Protein content of fresh seeds was also determined after harvest. Plants were harvested at the age of 135 days. Ten plants from each treatment were harvested separately to record data on different parameters. Data were analyzed statistically (Steel *et al.* 1997) and treatment means were compared by LSD test at 5% level of significance.

Results and Discussion

Results presented in Table 1 showed that application of different TIBA treatments had both stimulatory and retarding effect on plant height, number of tillers and leaves per plant in most of the cases. Here, application of 10 ppm TIBA although produced tallest plant and maximum number of leaves per plant but statistically at par with control. The present results are fairly in agreement with the findings of Islam (2014) on BRRI Dhan-44 and Munira (2015) on BARI Maize-6. In addition, application of TIBA treatments had retarding effects on plant height of chickpea (Islam and Jahan 2019) and stimulatory effect on number of branches and leaves per plant in mungbean (Adam and Jahan 2014) and chickpea (Islam and Jahan 2019). Seed treated with *Rhizobium* resulted significantly shortest plant, lowest number of tillers and leaves per plant. In chickpea, Jahan *et al.* (2018) also recorded the lowest number of primary branches per plant due to *Rhizobium* treatment although in case of height it was increased non-significantly. However,

results of Hoque and Haq (1994) on lentil and Togay *et al.* (2008) on chickpea reported that seed inoculated with *Rhizobium* significantly increased plant height.

Findings of this investigation indicated that dry weight of leaves and shoot decreased following TIBA and *Rhizobium* treatments but was significant due to *Rhizobium* inoculation. Increase in dry weight of root was obtained from 10 and 50 ppm TIBA treatments where, the maximum was due to 10 ppm TIBA application although statistically identical to control. Application of TIBA treatments had both increasing and decreasing effect on total dry matter per plant in BARI Mung-5 (Adam and Jahan 2014) and BARI Chola-7 (Islam and Jahan 2019). Seeds treated with *Rhizobium* also produced significantly lowest dry weight of root. This results are in agreement with the findings of Jahan *et al.* (2018) where they obtained lowest dry weight of shoot and root of chickpea from *Rhizobium* treatment. However, results of Ogutcu *et al.* (2010) had revealed that bacterial inoculations significantly increased dry weight of root in chickpea.

Leaf area ratio of BRRI Dhan-55 were recorded significantly higher due to all treatments and the maximum was noted from *Rhizobium* treated plants. Application of TIBA treatments produced higher LAR at 10 days after spray and then decreased gradually in BRRI Dhan-44 (Adam *et al.* 2015). Specific leaf weight of BRRI Dhan-55 was found to reduced following all treatments and was significant due to *Rhizobium* inoculation. Findings indicated that different concentrations of TIBA and *Rhizobium* inoculation had not produced any better results in relative growth rate (RGR) and net assimilation rate (NAR) of BRRI Dhan-55 and the minimum was observed from *Rhizobium* treated plants. Adam *et al.* (2015) recorded both stimulating and retarding effects of TIBA on SLW, RGR and NAR of BRRI Dhan-44 at different days after spray. The probable reason of decreasing RGR could be attributed to shading of lower leaves by upper leaves (Thorne 1961).

Application of TIBA and *Rhizobium* inoculum showed mostly retarding effects on yield attributes and yield of BRRI dhan-55 but with non-significant variations (Table 2). The maximum number of effective tillers per plant (15.10) was recorded from 10 ppm TIBA treatments although statistically at par to control and 50 ppm treatments. The only increase in number of non-effective tillers per plant was obtained from 50 ppm TIBA treatment but not significantly different from control and 10 ppm TIBA treatments. However, *Rhizobium* treated plants produced significantly lowest value in both the number of effective and non-effective tillers per plant. Results of this experiment is more or less similar to the findings of Adam *et al.* (2018) where, TIBA treatments had non-significant response on the number of effective and non-effective tillers per plant in BRRI Dhan-44. Outcome of Biswas *et al.* (2000) have showed increased number of panicles per plant of rice due to inoculation of rice seeds with *Rhizobium* sp. which is not in consistence with the findings of the present work.

Results revealed that length and dry weight of panicles per plant reduced following all treatments except 10 ppm TIBA and decrease was significant due to *Rhizobium* treatment only. Number of grains per plant was negatively influenced due to all treatments where all the treatments produced statistically identical value to control. Increase in % of filled grains per panicle was noted from 50 ppm TIBA only and was significantly higher than all other treatments. Results obtained in case of length of panicles, dry weight of panicles, number of grains per panicle and % of filled grain per panicle are in contrary with the findings of Adam *et al.* (2018) in BRRI Dhan-44. Findings in case of number of grains per plant are moderately in agreement with the outcome of Islam and Jahan (2019) in BARI Chola-7.

The highest per cent 38.47% of un-filled grains per panicle was found in *Rhizobium* treated plants followed by 10 and 25 ppm TIBA treatments respectively. However, plants of 50 ppm TIBA had

Table 1. Effects of TIBA and *Rhizobium* inoculation on growth parameters of BRRI Dhan-55 at harvest.

Treatments	Height (cm)	Number of tillers per plant	Number of leaves per plant	Dry weight of leaves (g)	Dry weight of shoot (g)	Dry weight of root (g)	Leaf area ratio (cm ² /g)	Specific leaf weight (g/cm ²)	Relative growth rate (g/g/day)	Net assimilation rate (g ² /day)
Control	128.06 a	15.70 a	103.80 a	12.43 a	44.22 a	14.51a	1.04 c	0.18 a	0.04	0.55
<i>Rhizobium</i> sp.	106.99 b	3.10 c	18.90 b	2.56 b	8.19 c	2.16 b	2.85 a	0.08 b	0.01	0.03
10 ppm TIBA	128.61 a	16.20 a	111.10 a	12.21 a	38.11 ab	15.50 a	1.05 c	0.17 a	0.05	0.15
25 ppm TIBA	121.00 a	12.20 b	86.90 a	8.99 a	23.03 bc	11.17 a	1.28 b	0.12 ab	0.03	0.13
50 ppm TIBA	128.50 a	16.30 a	94.90 a	12.22 a	35.99 a	15.40 a	1.29 b	0.15 a	0.04	0.16
CV (%)	13.82	18.54	25.53	11.75	15.21	16.63	50.60	13.96	10.13	14.29
LSD (0.05)	13.25	3.36	33.66	3.62	16.88	6.47	0.129	0.06	NS	NS

Mean in a vertical column followed by same letter or without letter do not differ significantly at 5 % level.

Table 2. Effects of TIBA and *Rhizobium* inoculation on yield attributes and yield of BRRI Dhan-55 at harvest.

Treatments	No. of effective tillers/plant	No. of non-effective tillers/plant	Length of panicle (cm)	Dry weight of panicle/plant (g)	No. of grains/panicle	% of filled grains	% of unfilled grains	1000-seed weight (g)	Yield /plant (g)	Harvest index (%)
Control	14.50ab	1.20ab	24.43a	44.09a	135.57	76.42b	23.58c	22.93bc	698.60a	36.28b
<i>Rhizobium</i> sp.	3.10c	0.00c	23.27b	12.75b	121.16	61.53b	38.47a	28.04a	33.93c	46.90a
10 ppm TIBA	15.10a	1.10ab	24.45a	44.14a	127.04	65.15b	34.86ab	24.11b	715.90a	38.39a
25 ppm TIBA	11.30b	0.90b	24.42a	36.48a	135.54	71.59b	28.41bc	24.59b	404.86b	37.68a
50 ppm TIBA	14.90ab	1.40a	24.11ab	42.02a	126.17	86.19a	13.81d	21.20c	557.57ab	38.61a
CV (%)	6.69	12.84	4.02	4.29	14.11	14.24	6.93	11.86	7.86	22.57
LSD (0.05)	3.73	0.4	1.08	10.46	NS	6.91	6.91	2.21	293.70	9.23

Mean in a vertical column followed by same letter or without letter do not differ significantly at 5 % level.

Table 3. Effects of TIBA and *Rhizobium* inoculation on pigment content of leaves of BRRI Dhan-55 at three different stages.

	Tillering stage			Flowering stage			Grain filling stage		
	Chl. a	Chl. b	Carotenoids	Chl. a	Chl. b	Carotenoids	Chl. a	Chl. b	Carotenoids
Control	1.06 b	0.46 b	7.58	1.44 a	0.87 a	10.72	0.16 a	0.47 a	2.52 b
<i>Rhizobium</i> sp.	1.29 b	0.50 b	7.48	0.62 b	0.43 b	8.94	0.08 b	0.10 c	5.33 a
10 ppm TIBA	1.81 a	0.80 a	8.55	0.44 b	0.82 a	10.07	0.08 b	0.10 c	3.23 a
25 ppm TIBA	0.41 c	0.51 b	9.05	0.53 b	0.98 ab	9.66	0.19 a	0.25 b	5.84 a
50 ppm TIBA	0.29 c	0.33 b	8.56	0.61 b	1.07 a	10.58	0.16 a	0.15 bc	4.94 a
CV (%)	16.23	16.49	13.03	16.25	13.84	11.74	12.57	7.32	14.22
LSD (0.05)	0.33	0.23	NS	0.20	0.59	NS	0.07	0.10	1.31

Mean in a vertical column followed by same letter or without letter do not differ significantly at 5% level.

produced significantly lowest % of un-filled grains per panicle. Application of TIBA treatments had both inducing and retarding responses on % of un-filled grains per panicle but with non-significant variations (Adam *et al.* 2018). Oladele and Awodun (2014) reported that inoculation of seeds by *Rhizobium* sp. produced longest panicle, maximum number of grains and un-filled grains per panicle in rice plants which is partially in agreement to the findings of present investigation.

Findings presented in Table 2 indicated that *Rhizobium* treated plants resulted significantly maximum 1000-seed weight whereas, application of 10 and 25 ppm although produced significantly higher 1000-seed weight than 50 ppm TIBA but statistically similar to control. Outcome of Hussain *et al.* (2009) revealed that *Rhizobium* sp. had produced significantly higher 1000-grain weight of rice than control treatment. By applying TIBA, similar results of increase in 100-seed weight of BARI Chola-7 was noted by Islam and Jahan (2019). Yield per plant of BRRI Dhan-55 was slightly increased following 10 ppm TIBA treatment although statistically identical to control and 50 ppm TIBA applications. Increase in yield per plant of BRRI Dhan-55 due to 10 ppm TIBA was 2.48% over the control. Results also showed that *Rhizobium* treated plants produced significantly lowest amount of yield. Foliar application of TIBA had resulted significantly higher yield per plant in BRRI Dhan-44 (Adam *et al.* 2018). However, Islam and Jahan (2019) recorded significantly higher yield per plant due to 10 and 20 ppm TIBA treatment but concentrations above 20 ppm decreased yield non-significantly.

Significantly higher harvest index were recorded following all treatments where the maximum was obtained from *Rhizobium* treated plants followed by 50, 10 and 25 ppm TIBA respectively. Previous investigations revealed that TIBA treatments resulted higher harvest index in various crops but with similar statistical values (Adam *et al.* 2018, Islam and Jahan 2019). The most probable reason for obtaining maximum harvest index in *Rhizobium* treated plants might be due to the production of higher 1000-seed weight which ultimately increased economic yield.

Pigment contents of leaves decreased with few exceptions due to application of TIBA and *Rhizobium* inoculum (Table 3). Significantly higher chl. a and chl. b was noted from 25 ppm TIBA at tillering stage whereas, at grain filling stage, all treatments produced significantly higher carotenoids content than control. Results also revealed that the effect of magnitude of TIBA were different depending on the concentration and stage of plant growth. Findings on pigment content of leaves are in accord with the results of Munira (2015) in maize and Adam *et al.* (2018) in rice where, they have observed that pigment content of TIBA treated plants may increase or decrease depending on the concentrations used and also on the plant.

Table 4. Effects of TIBA and *Rhizobium* inoculation on protein content of leaves and seeds of BRRI Dhan-55 at different stages.

Treatments	Protein content of leaves(mg/g)			Protein content of seeds (mg/g)
	Tillering stage	Flowering stage	Grain filling stage	At harvest
Control	465.63 a	18.88 b	28.56 a	10.18 c
<i>Rhizobium</i> sp.	225.00 c	12.94 c	15.65 c	5.05 d
10 ppm TIBA	311.00 bc	27.63 a	24.04 ab	13.00 b
25 ppm TIBA	315.88 b	25.88 a	22.83 abc	18.25 a
50 ppm TIBA	379.75 ab	21.40 b	17.08 b	5.11 d
CV (%)	25.78	26.34	26.72	8.11
LSD (0.05)	86.22	4.26	7.51	1.84

Mean in a vertical column followed by same letter or without letter do not differ significantly at 5 % level.

Results presented in Table 4 showed that TIBA treatments had significant positive effects on protein content of leaves at flowering stage. However, the influence was rather negative at both tillering and grain filling stages. Foliar application of TIBA had affirmative response on protein content of leaves at tillering stage and negative effects at flowering and grain filling stages (Adam *et al.* 2018). Results also showed that at harvest, significantly highest protein content of seeds (18.25 mg/g) was obtained from 25 ppm followed by 10 ppm TIBA treatment (13.00 mg/g). The least protein content of seeds (5.05 mg/g) was observed from *Rhizobium* treated plants which was statistically identical to 50 ppm TIBA but significantly lower than rest of the treatments. The present findings are also in agreement with the findings of Munira (2015).

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