EFFECT OF FOLIAR APPLICATION OF SALICYLIC ACID ON YIELD AND YIELD CONTRIBUTING CHARACTERS OF BRRI hybrid dhan3

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Dedicated

To

My Beloved

Parents

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CERTIFICATE

*This is to certify that thesis entitled "***EFFECT OF FOLIAR APPLICATION OF SALICYLIC ACID ON YIELD AND YIELD CONTRIBUTING CHARACTERS OF BRRI hybrid dhan3"***submitted to the Department of Soil Science, Sher-e-Bangla Agricultural University (SAU), Dhaka, in partial fulfillment of the requirements for the degree of* ***MASTER OF SCIENCE (MS) IN SOIL SCIENCE****, embodies the result of a piece of bonafide research work carried out by* ***MOST.MOSLAMA KHATUN,*** *Registration no.***14-06322** *under my supervision and guidance. No part of the thesis has been submitted earlier for any other degree or diploma.*

I further certify that such help or source of information, as has been availed of during the course of this investigation has duly been acknowledged.

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SAU, Dhaka, Bangladesh ***The Author***

EFFECT OF FOLIAR APPLICATION OF SALICYLIC ACID ON GROWTH AND YIELD OF BRRI hybrid dhan3

ABSTRACT

The experiment was conducted in the Farm of Sher-e-Bangla Agricultural University, Dhaka, Bangladesh during the period from November 2014 - April 2015 to study the effect of different levels of salicylic acid (SA) on growth and yield of BRRI hybrid dhan3.The experiment included six levels of SA viz. T_1= 0, T_2= 200, T_3= 400, T_4= 600, T_5= 800 and T_6= 1000 µM. The experiment was laid out in a randomized complete block design with three replications. The results revealed that, Lower dose of SA has a positive role on plant height of rice and higher doses up to 1000 µM have negative effect. The highest oven dry matter weight (2.7 t/ha) was found in T_3 treatment having foliar spray of SA at 400 µM and the lowest (2.0 t/ha) was found in, T_5. The highest number of effective tillers per hill (14) was found in the treatment T_6 due to the application of 1000 µM SA and the highest 1000 grain weight (34.8g) was found with the same treatment. The highest number of grain per panicle (156) was found in the T_6 treatment having foliar application of 1000 µM SA and it was statistically similar with the treatments T_4 and T_5. The application of different levels of SA had a significant effect on number of stem borer affected plants. The lowest number of affected panicle per plot (3) was found in the treatment T_6. The maximum grain yield was found in the treatment T_6 having 1000 µM SA and it was statistically identical with the treatment T_5 having 800 µM SA. On the other hand, minimum grain yield was found in the control treatment T_1 which was similar to the treatments T_2 and T_3 having 200 µM and 400 µM SA, respectively. So, it can be concluded that salicylic acid may be applied as foliar application to rice crops for better growth and yield.

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LIST OF ABBREVIATIONS

AEZ	=	Agro Ecological Zone
BARI	=	Bangladesh Agricultural Research Institute
BAU	=	Bangladesh Agricultural University
BSS	=	Bangladesh Bureau of statistics
cm	=	Centimeter
^0C	=	Degree Celsius
DAT	=	Days after transplanting
DMRT	=	Duncan's Multiple Range Test
et al.	=	and others
FAO	=	Food and Agriculture Organization
Fig.	=	Figure
g	=	gram (s)
HI	=	Harvest Index
kg	=	Kilogram
kg/ha	=	Kilogram/hectare
LER	=	Land Equivalent Ratio
LSD	=	Least Significant Difference
SE	=	Standard Error
CV	=	Co Efficient of Variation
µM	=	Micro Mole
MoP	=	Muriate of Potash
m	=	Meter

pH	=	Hydrogen ion conc.
ppm	=	Parts Per Million
RCBD	=	Randomized Complete Block Design
SA	=	Salicylic acid
SAU	=	Sher-e-Bangla Agricultural University
TSP	=	Triple Super Phosphate
t/ha	=	ton/hectare
UNDP	=	United Nations Development Programme
%	=	Percent
Wt.	=	Weight

INTRODUCTION

Chapter 1

INTRODUCTION

Bangladesh is an agro-based country. Rice belongs to the Gramineae family with the genus *Oryza* which contained about 22 different species (Wopereis *et al*., 2009). It is the dominant staple food for many countries of the world (Mobasser *et al*., 2007). It is also the most important food crop and a major food grain for more than a third of the world and 50% of the global population (Zhao *et al*., 2011). Among the most cultivated cereals in the world, rice ranks at second to wheat (Abodolereza and Racionzer, 2009). Rice is grown in more than 10 countries with a total harvested area of nearly 160million hectares, producing more than 700million tons every year (IRRI, 2010). According to the FAO, of the UN, 80% of the world rice production comes from 7 countries (UAE/FAO, 2012).

In Bangladesh, rice covered an area of 28.49million acres with a production of 33.54 million tons while the average yield of rice in Bangladesh is around 1.18 tons/acre (BBS2012). In case of Boro rice, it covers the largest area of 11788 (41.4% of total rice cultivation area) acre (local 195 + HYV 9968 + HYV 1625 acre) with a production of 1.86 million tons (55.5%) and the average yield is about 1177 kg/acre during 2010-2011 (BBS, 2012). Besides, based on the rice cultivation, Bangladesh is the 4th largest country of the world (BRRI, 2016). Alam (2012) also reported that rice covers about 82% of the total cropped land of Bangladesh. It accounts for 92% of the total food grain production in the country and provides more than 50% of the agricultural value addition employing about 44% of total labour forces. According to the latest estimation made by BBS, per capita rice consumption is about 166 kg/year. Rice alone provides 76% of the calorie intake and 66% of the total protein requirement and shares about 95% of the total cereal food supply (Alam, 2012).

The population of Bangladesh is growing by two million every year and increase by another 30 million over the next 20 years. Thus, Bangladesh will require about 27.26 million tons of rice for the year 2020 (BRRI, 2011). During this time total rice area will also shrink to 10.28 million hectares. Rice yield therefore, needs to be increased by 53.3% (Mahamud*et al.*, 2013). In Bangladesh, food security has been and will remain a major concern because food requirement is increasing at an alarming rate due to increasing population. Rice yield, in general, is comparatively lower than that of other South east Asian countries because of severe insect infestation, drought, salinity etc. Yield loss up to 50% has been recorded in susceptible rice varieties when all the leaf sheaths and leaf blades were infected (Kumar *et al.*, 1999). The crop is suffering from different fungal, bacterial, viral and mycoplasmal diseases. However, in order to defend themselves against these attacks, plants have evolved various constitutive and inducible mechanisms, one such mechanism being the accumulation of large quantities of salicylic acid. Salicylic acid (SA), a phenolic compound, is associated with stress tolerance in plants. Previous studies reported that SA can induce tolerance against high and low temperatures, drought, salinity, ultraviolet light, heavy metal toxicity, diseases and pathogens (Raskin, 1992; Yalpani *et al*., 1994; Dat *et al*., 1998; Metwally *et al*., 2003; Sakhabutdinova *et al*., 2003; Hayat & Ahmad, 2007; Horváth *et al*., 2007, Farooq *et al*., 2008b; Hussain *et al*., 2008). In addition, SA plays an essential role in preventing oxidative damage in plants by detoxifying superoxideradicals (Bowler *et al*., 1992) and is also involved in calcium signaling (Kawano *et al*., 1998). Plants treated with SA showed increased vigor of early seedling growth (Farooq *et al*., 2008), increased photosynthetic rates and water use efficiency, decreased stomatal conductance and transpiration rate (Khan *et al*., 2010). Moreover, there is evidence that exogenous application of SA can alter antioxidant capacity in plants (Rao *et al*., 1997), providing protection against oxidative damage (Larkindale & Huang 2004), and thereby inducing stress tolerance.

In this study, SA is important in plant growth and development and is also associated with stress tolerance in plants. Plants pre-treated with SA (Larkindale & Knight, 2002) showed induced stress tolerance and protection against oxidative damage due to various stresses. Despite the importance of these chemicals in stress tolerance, little is known about their effects on rice morphology, phenology and physiology. The research presented herein addresses the effects of SA on rice morphology, phenology and physiology, under non-stress conditions. Thus, studies that seek this information, besides aiming to learn which products can be used to stimulate grain yield and yield contributing characters, should be encouraged. Within this context, the aim of this present study was carried out

- To examine the performance of hybrid rice with foliar application of SA.
- To evaluate the foliar application doses of SA for the yield management of BRRI hybrid dhan3.

REVIEW OF LITERATURE

Chapter 2

REVIEW OF LITERATURE

Rice (*Oryza sativa*) is the staple food crops in Bangladesh. Growth and development of rice plant are greatly influenced by the environmental factors, variety and cultural practices. Among these factors, irrigation and SA play notable role regarding the growth and development of rice plants. A good number of research works have been conducted at home and abroad on the effect of foliar application of SA on cultivation.

SA is more or less unknown to the farmers in producing crops in Bangladesh. The present study deals with effect of foliar application of SA on productivity of rice. For aiming this point, foliar application of SA used to evaluate the different roles of morpho-physiology, yield and yield attributing characters of BRRI hybrid dhan3 at SAU campus. In this case an attempt is made to review the available literature pertaining to the present study. Most of the research reports showed a positive effect of the foliar application of SA on yield of rice and other crops. The findings of various authors are cited below:

2.1 Application of Salicylic Acid on Rice

Singh, *et al.*(2015) carried out an experiment to assess the effect of salicylic acid along with standard fungicide on sheath infecting pathogen and yield attributes in hybrid rice. Two different concentration of SA (20 and 40 ppm) and Mancozeb (3 and 4 g/L) were used at three different stages (Booting stage, Heading stage and at the time of 50% flowering). Results revealed significant increase for most of the yield attributes studied for all the treatments over control but found non-significant for panicle/plant. The Area under

Diseased Progress Curved (AUDPC) was decreased significantly for all the treatments over control. The correlation between AUDPC and yield parameters was varied. AUDPC was strongly correlated with all the yield attributes. The value of AUDPC was negatively correlated to different yield attributes proving that the pathogen had a damaging effect on the yield attributes of hybrid rice.

Tavares, *et al.* (2014) showed that Seed treatment with growth regulators, especially SA is a promising alternative to the seed industry because it is an important inducer of resistance to diseases and pests, as well as acting significantly on quality and seed yield. The treatments consisted of increasing levels of 0, 50, 100, 150 and 200 mg/L SA. The physiological quality of seeds produced was treated and evaluated by tests of vigor and germination, and after harvest were evaluated seed yield. It follows that treatment of rice seeds with SA concentrations up to 130 mg/L at a solution dose of 2 ml/kg seed does not affect the germination and affects the strength, however provides substantial increases in the yield of seeds. The seed treatment with SA has no influence on seed quality produced.

Usharani, *et al.*(2014) showed that, the effect of SA and *Pseudomonas fluoresces* on growth and yield of Paddy IR-50. Among the various treatments tested, maximum growth and yield was observed in the treatment (*Pseudomonas fluorescens*6 seed application + SA applied on 30th day) and the least parameters were recorded in the Control treatment.

Ghodrat, *et al.*(2013) revealed that plant growth and development are controlled by plant growth regulators and pre-sowing seed treatments have proven advantageous to germination, seedling growth and yield of several field crops, such as rice (*Oryza sativa* L.) cultivar Sadri to soaking seeds for three days in 0.0, 50 and 100 mg/Lindole butyric acid (IBA), gibberellic acid (GA) and SA (SA) and results showed that percent germination and germination rate were significantly higher in all the hormonal treatments. Root and shoot lengths were increased significantly by nearly all the treatments. The fresh and dry weights at seedlings were affected the most by IBA. Significant increase in chlorophyll, carotenoids and anthocyanin contents of leaves occurred by soaking seeds in hormonal solutions. Except for IBA, treatments did not change tillers number significantly. Panicle grain numbers increased significantly by all the treatments, whereas, 1000 kernels weight was affected only by IBA and SA at 50 mg/L. IBA @ 50 mg/L had the highest impact on grain yield with 9.4 compared to 7.5 t/ha in control which is more than 25% increase in grain yield. Among the treatments, IBA @ 50 mg/L significantly affected all the yield tested components.

Sood, *et al.* (2013) showed that the effects of elicitors *viz*. benzothiadiazole (BTH) and SA on defense related enzymes *viz.* peroxides, phenylalanine ammonia lyase, superoxide dismutase, chitinase and β-1,3-glucanase, and phenols in rice (Pusa Basmati I) plants. First foliar spray of BTH (50 mg/kg) and SA (50 mg/kg) was done at the maximum tillering stage and inoculation with *Rhizoctoniasolani* was carried 24 h after elicitor treatment. Elicitors were further sprayed at every growth stage. Time course analysis showed peak accumulation of defense related enzymes and phenols in the rice leaves treated with BTH and SA, and accumulation was the highest at the flowering stage. Higher enzymatic activity was observed in elicitor treated plants inoculated with *R. solani*. Compared to the untreated control plants, application of elicitors before *R. solani* inoculation significantly elicited the defense related enzymes and phenols. Moreover, application of elicitors had a positive effect on yield and disease reduction. Mohammed, R.A. (2011) carried out an experiment where the impacts of α-tocopherol, glycine betaine (GB) and SA applications on higher plants have been the subject of many studies, with special emphasis on oxidative stress tolerance under adverse conditions. However, little work has been carried out on rice responses to α-tocopherol, GB or SA under non-stress conditions, in which yield could potentially increase. This study determined the effects of α-tocopherol (2.3 kg/ha), GB (2.0 kg/ha) or SA (12.9 kg/ha) application on rice morphology, phenology and physiology undernon-stress conditions. The applications did not affect production of tillers, biomass, phenology, or pollen germination; however, plant height, leaf characteristics and physiology, spikelet fertility (SF), panicle and grain characteristics and yield were affected. Plants treated with α-tocopherol, GB or SA showed 6%, 13% and 13.5%increases in grain yield as a result of decreased respiration and increased membrane integrity and SF.

Farooq, *et al.* (2009)showed that the role of salicylic acid (SA) to induce drought tolerance in aromatic fine grain rice cultivar Basmati 2000 was evaluated. SA was applied as seed and foliar treatments. For seed treatment, rice seeds were soaked in 50, 100 and150 mg/L aerated solution of SA for 48 h and then dried back. Treated and untreated seeds were sown in plastic pots in a phytotron. At four leaf stage, one set of plants was subjected to drought stress, while the other remained well watered. Drought was maintained at 50% of field capacity by watering every alternate day. For exogenous application, SA was applied 50, 100 and 150 mg/L at five leaf stage. In the control, SA was neither applied exogenously nor as seed treatment. Drought stress severely affected the seedling fresh and dry weight, photosynthesis, stomatal conductance, plant water relations and starch metabolism; however, SA application improved the performance of rice under both normal and stress conditions. Drought tolerance in rice was well associated with the accumulation of compatible solutes, maintenance of tissue water potential and enhanced potency of antioxidant system, which improved the integrity of cellular membranes and facilitated the rice plant to sustain photosynthesis and general metabolism. Foliar treatments were more effective than the seed treatments. Foliar application with 100 mg/L (FA 100) was the best treatment to induce the drought tolerance and improve the performance under normal and stress conditions compared with the control or other treatments used in this study.

2.2 Application of Salicylic acid on Others Crop:

Amira M. S. Abdul Qados (2015) carried out an experiment to investigate the effect of salinity stress on growth, chemical constituents and yield, and to examine whether salinity stress can be offset by the exogenous application of SA on sweet pepper (*Capsicum annuum L. cv. Orlando*). Salinity stress (2000, 4000 or 6000 ppm) decreased plant growth and marketable yield but SA (250 ppm) treatment as foliar spray counteracted significantly the harmful effects of low and moderate salinity stress levels (2000 and 4000 ppm) and partially counteracted the harmful effects under the highest salinity stress level (6000 ppm).

Desoky & Merwad (2015) conducted a pot experiment to evaluate the response of wheat plants (*Triticum aestivum* L.) cv. Sakha 93 to different levels of foliar spray of some antioxidant tested substances as ascorbic and SA at a rate of 0.1 and 0.2%, with respect to vegetative criteria, some physiological properties *i.e.* phenol components, proline concentration, yield components, NPK-uptake as well as anatomical structure of flag leaf blade grown under salt stress conditions, 3.21 dSm^{-1} , 6.32 dSm^{-1} and 10.65 dSm^{-1} of soil salt. Data indicated that, all studied vegetative criteria of wheat plants, decreased under salt stress condition. Spraying antioxidant substances seemed to partially overcome the harmful effects of salt stress on vegetative criteria. The highest values of straw and grain yield, biological yield, weight of 1000 grain, protein content and yield efficiency, straw and grains N, P and K-uptake of wheat plants occurred with ascorbic acid 0.2% "AA_2" treatment followed by ascorbic acid 0.1% "AA_1", SA 0.2% "SA_2", salicylic acid 0.1% "SA_1" and untreated plants that descending order in the two seasons.

Ahmad *et al.* (2014) conducted an experiment in pots and field to investigate the effect of exogenous application of ascorbic acid (AsA), SA and $H_2O_2$to improve the maize performance at sub-optimum temperatures. In pot experiment, foliar application of AsA, SA and H_2O_2 at each concentration improved seedling growth, leaf relative water, chlorophyll *b* contents, membrane stability and enzymatic antioxidant activities in maize. In field experiment, application of these substances either through seed priming or foliar spray improved the morphological, yield related attributes and grain yield of spring maize; however, seed priming was more effective than foliar application.

Molazem, D.& Bashirzadeh, A. (2014) conducted an experiment to evaluate the effect of salt stress and salicylic acid application on growth and physiological traits of maize varieties in research farm of Islamic Azad University of Ardebil branch during 2012-13.Salt stress factor including three levels (control, 50mM and 100mMNaCl) and SA (control, 1mM and 2mM). Results from the experiment showed that, between different salinity in carotenoid, chlorophyll a+b, chlorophyll content and proline were significantly different. Effect of SA except for stem diameter was not significant for all traits. Between leaves length with chlorophyll content, total chlorophyll (a+b) and carotenoid was observed significant positive correlation. There was a significant positive correlation between chlorophyll content with total chlorophyll, carotenoids and stem diameter.

Mahdi, J.(2014) revealed that the effects of SA on some quality characters of tomato different concentration of SA (10^{-2}, 10^{-4}, 10^{-6}, 10^{-8} molar and control) was done in seedling stage as foliar replication. Measured characters was including (number of panicle in a bush, yield, fruit number in panicle, fruit number in bush, fruit weight and fruit diameter). Obtained results of this study show that SA significantly affected number of panicle in a bush, yield, fruit number in panicle, fruit number in bush, fruit weight and fruit diameter. Among foliar application, the highest rate of tomato yield with mean of 3059.5 g obtained in SA_3 (SA at 10^{-6} M), highest numbers of panicle in tomato bushes with mean of 31.25 measured in SA_1 (SA at 10^{-2} M). The highest fruit number in panicle and highest fruit number in bush obtained by mean of 3.5 and 66.75 in SA_1 (SA at 10^{-2} M), respectively and minimum amount of all this characters was recorded in control treatment and the highest amount of fruit weight and also fruit diameter was measured in control treatment with mean of 61.50 g and 51.75 mm, respectively.

Ibrahim *et al.*(2014) carried out two cemented plots experiments during the winter seasons of 2012-13 and 2013-14, Soil Salinity Laboratory, Alexandria, Egypt, to study the effect of three levels of SA (0, 50, 100 ppm) on yield and yield components of wheat (Sakha 93). Increasing SA rates resulted in significant increase in plant height (cm), number of grain/spike, number of spikes/m^2, 1000 grain weight (g), grain yield (g/plot), straw yield (g/plot), and biological yield (g/plot) in addition to grain weight/spike (g).

Howladar & Dennett (2014) conducted a pot experiment to analyze the effect of exogenous application; seed priming or foliar spraying of SA on YecoraRojo and Paragon wheat cv. under NaCl-salinity. Gas exchange parameters, growth parameters, yield and yield components were reduced in both cultivars under salinity stress with foliar spray and soaking seeds. Exogenous application of SA through foliar spraying or seed soaking showed a slight increases or decreases with the application method or between cultivars. SA foliar spraying exhibited a slight improvement over SA seed soaking in most parameters, particularly in Paragon. However, the low SA concentration; 0.5 mM tended to improve most parameters in both cultivars.

Morad *et al.* (2013) showed that the effect of salt stress and SA application on growth and yield component traits of wheat in research farm (green house condition) of university of tehran (karaj-iran) during 2010-11. Salt stress factor including three levels (control, salt stress with NaCl (4 and 8 ds/m) and SA (application and none application). The experiment was carried out on two variety of wheat, separately. The results indicated that maximum height was achieved in Control × SA none application treatment and minimum height was achieved in NaCl 8 ds/m × SA none application treatment. SA application increased number of grain in spike. SA application alleviated destructive effect of salt stress. The results indicated that interaction effect of salt stress × SA had significant effect ($p \leq 0.01$) on Tabasi variety but had not significant effect on Arv and variety on total chlorophyll and relative water content traits. It can be concluded that foliar application of wheat cultivar plants with SA stimulate the growth of wheat plants via the enhancement of the biosynthesis of photosynthetic pigments; improved relative water content, decreasing of organic solutes (Proline) and thus SA treatment improved wheat growth especially on Tabasi variety.

Javed*et al.* (2013)showed that the effect of SA on growth and nitrogen metabolism in mungbean grown under saline conditions, an experiment was conducted in wire house in plastic pots containing soil + sand at NIAB Faisalabd. Mungbean (*Vigna radiate* L.) varieties, two salt tolerant (NM-98 and NM-92) and two salt sensitive (NM-54 and NM-13-1), identified in laboratory experiments, were grown under four salinity levels, i.e., 1.2, 4, 8 and 12 dS/m. Salicylic acid @ 0, 100, 200 and 300 mg/L was applied as foliar spray at vegetative and flowering stages. Results indicated that salinity reduced the growth by decreasing plant height and fresh biomass of all the cultivars, however, the salt tolerant cultivars performed better than sensitive ones. Foliar application of SA @ 100 mg/L significantly improved all the growth parameters in all the cultivars under saline conditions. The SA levels of 200 and 300 mg/L did not show appreciable performance regarding growth attributes under normal and saline conditions. Application of SA @ 100 mg/L was helpful in reducing the adverse effects of salinity on all the above mentioned parameters while other levels of SA did not perform better under all salinity treatments.

Ghasemi-Fasaei, R. (2013) showed that foliar application of SA decreased mean dry weight of cucumber by 31%, while its effect on mean dry weight of chickpea was negligible. Foliar application of SA decreased mean uptake of Zn, Mn, Cu and Fe in cucumber shoot by 29, 34, 22 and 31%, respectively. Decrease in dry matter yield of cucumber following foliar application of SA was, therefore, attributed to the significant decrease in the uptakes of metal micro nutrients. Foliar application of SA decreased mean uptake of Cu and Fe in chickpea shoot by 31 and 18%, respectively. The effect of SA on mean Zn uptake in chickpea shoot was negligible. Foliar application of SA caused an increase in mean Mn uptake of chickpea shoot by about 7%.The influence of HA levels on mean dry matter weight in chickpea was uncertain. Soil application of 2 and 4 mg HA kg^{-1} caused negligible decrease in mean dry matter weight of chickpea shoot by 5 and 8%, respectively. The effects of HA levels on the uptakes of Mn was insignificant. Application of 4 mg HA/kg increased mean uptakes of Fe, Cu and Zn by 7.1, 8.5 and 9.6%, respectively. Application of 4 mg HA/kg increased mean uptakes of metal micronutrients compared to the control although the increase for Fe uptake was negligible. Application of 4 mg HA/kg increased mean uptakes of Mn, Cu and Zn by 18.7, 100, and 18.6%, respectively. Shoot dry weight of chickpea was significantly correlated with the uptakes of

Zn, Fe and Mn but was not correlated with the uptake of Cu. Shoot dry weight of cucumber was significantly correlated with the uptakes of Zn, Fe, Cu and Mn. In cucumber shoot, the uptakes of all metal micronutrients were significantly correlated with each other. In chickpea shoot, the uptakes of all metal micronutrients other than Fe were significantly correlated with each other. According to the results of present study it appears that neither SA nor HA was efficient to be recommendable for correcting metal micronutrients deficiency under micronutrients deficient conditions.

Salwa *et al.* (2013) conducted the work concerned to study the effect of SA on growth criteria (shoot height and shoot dry weight), soluble sugars and protein, antioxidant enzymes (SOD, APX and GR) activities and specific activities, lipid peroxidation, electrolyte leakage and yield criteria (Pod weight, seed weight, seed number and 100-seed wieght). The obtained results revealed that salt treatments provoked oxidative stress in faba bean plants as shown by the increase in lipid per oxidation and electrolyte leakage and consequently negatively affected growth and yield criteria. Foliar spray with SA at the concentration of 2mM followed by 1mM mitigated the harmful effects of salt stress through the enhancement of the protective parameters, such as antioxidant enzymes, soluble sugars and proteins and consequently improved growth and yield criteria.

Fahad S.& Asgharibano (2012) conducted a field experiment to determine the effect of exogenously applied SA on physiology of maize (*Zea mays* L.) hybrid cv. 3025 grown in saline field (pH 8.4 and EC 4.2 dS/m) as well as on the nutrient status of saline soil. The salicylic acid (10^{-5} M) was applied as foliar spray, 40 days after sowing (DAS) at vegetative stage of maize plants. Foliar application of SA to salt stressed plants further augmented the sugar, protein, proline, superoxide dismutase (SOD), peroxidase dismutase

(POD) ascorbate peroxidase (APOX) activities, endogenous abscisic acid (ABA) , indole acetic acid (IAA) content, and root length, fresh and dry weights of roots whereas, the chlorophyll a/b and ABA/IAA ratio were decreased. The exogenous application of SA significantly decreased the Na^{+}, Ni^{+3}, Pb^{+4}, Zn^{+2}, and Na^{+}/K^{+} content of soil and roots while increased the Co^{+3}, Mn^{+2}, Cu^{+3}, Fe^{+2}, K^{+} and Mg^{+2} content under salinity stress. It can be inferred that exogenous application of SA (10^{-5}M) was effective in ameliorating the adverse effects of salinity on nutrient status of soil. SA (10^{-5}M) can be implicated to mitigate the adverse effects of salinity on maize plants.

Rao *et al.* (2012) conducted a pot experiment to determine drought mitigating effect of SA and L-Tryptophan. Salicylic acid and L Tryptophan were sprayed at 3-4 leaves stage @ 100, 150, 200 ppm and 5, 10, 15 ppm, respectively. Drought stress was induced by withholding water after five days of SA and L-Tryptophan application. Significantly higher relative water content, leaf membrane stability index, chlorophyll and potassium content were found in plants treated with 100 ppm Salicylic acid and 15 ppm L-Tryptophan compared with other treatments and control plants. Results suggest that foliar application of SA and L-Tryptophan can play a role to reduce the effect of drought in maize.

Pradeep, et al. (2012) showed that four chickpea genotypes (Tyson, ICC 4958, JG 315 and DCP 92-3) were treated with 1.0 mM and 1.5 Mm SA and subjected to pre and post flowering drought stress to analyse its influence on nitrate reductase (NR) activity, relative water content (RWC), proline and antioxidant enzymes activity (superoxide dismutase and peroxidase). Leaf RWC significantly reduced during stress at both the growth stages and ranged between 71.7-74.4% (unstressed) and 68.0-71.7% (stressed), whereas in 1.5 mM

SA treated plants leaf RWC increased comparable to the control (unstressed plant). NR activity significantly reduced under stress at the post anthesis stage of growth but was maintained higher in 1.5 mM SA treated plants in all the four genotypes studied. On the other hand, activities of antioxidant enzymes superoxide dismutase (SOD) and peroxidase(POX) were up regulated by drought stress and interestingly further enhanced by 1.5 mM SA treatment. The response of SA (1.5 mM) was relatively more in ICC 4958 and Tyson cultivars of chickpea. Hence, results signify the role of SA in protecting metabolic activity along with regulating the drought response of plants.

Sadeghipour, O. & Aghaei, P. (2012) conducted an experiment to evaluate the influence of exogenous SA application on some traits of common bean under water stress conditions in Iran during 2011. Results showed that drought decreased plant height, leaf area index (LAI) and protein yield but increased seed protein content. Nevertheless, seeds soaking in SA (especially 0.5 mM) diminished drought damages and increased plant height, LAI and protein yield in both water stress and optimum conditions. SA application also decreased seed protein content. Results indicate that exogenous application of this phytohormone can act as an effective tool in improving the growth and production of common bean under water stress conditions.

Hassanein *et al.* (2012) conducted an experiment to study the analysis of drought stress defense triggers in wheat plants grown in dry sandy lands using methods of grain-priming and/or foliar pretreatments on the preanthesis stages. Morphological, biochemical and yield components data revealed that wheat originated from grain-priming combined with foliar applications had exhibited stronger anti-drought effects. A raised tolerance level was ascertained from the up-regulation of crop production and quality in drought cultivation compared to normally irrigated wheat.

Fateh *et al.* (2012) conducted an experiment to study the effect of SA and seed weight on Wheat germination (CV. BC ROSHAN) under different levels of osmotic stress. The results showed that osmotic stress decreased seed germination of wheat cultivars in general concentration of PEG (12 bar) and 1000 weight kernel (22 g) decreased germination over % as compared with control. Also, the SA increased the seedling length and dry weight of seedlings. SA increased length and weight of radicle and plumule in treatments of low seed weight (1000 grain weight =22 g).The lowest germination index were also observed in the treatment of severe stress and without pre-treated with SA and minimum seed weight.

Sharafizad *et al.* (2012) conducted an experiment to investigate the effect of SA on total yield and yield component of wheat under stress condition. Treatments were drought stress at three levels (control, drought stress in mid florescence and drought stress in grain filling stage). Second treatment was application of SA as a priming agent, foliar application at beginning of tillering and flowering, and the third treatment was different dosage of salicylic acid (0, 0.7, 1.2 and 2.7mM). Results of experiment showed that drought stress significantly decreased grain yield, efficiency of material distribution while the highest grain yield was obtained at non-stressed condition with application of 0.7mMSA. Grain yield exhibited high and positive correlation with number of spikes in m2, number of grain in spike, biological yield and harvest index.

Anosheh *et al.* (2012) carried out a field experiment to assess the effects of SA on drought-stress induced changes in morpho-physiological and biochemical characteristics of two commonly grown wheat cultivars in Iran. Drought stress increased canopy temperature and decreased leaf area index and plant height in both cultivars; however, exogenous applications of SA (0.7 mM) reduced these harmful effects considerably. Drought stress also significantly increased the levels of total soluble proteins and free proline, the activities of antioxidant enzymes superoxide dismutase, peroxidase and catalase, and decreased the contents of chlorophyll *a* and chlorophyll *b*. Application of SA increased total soluble proteins, chlorophylls *a* and *b*, and peroxidase activity.

El-Yazied, A.A. (2011) conducted a field experiment to study the effect of foliar application with 50 & 100 ppm of SA and 50 & 100 ppm chelated zinc (Zn) and their combination on some growth aspects, photosynthetic pigments, minerals, endogenous phytohormones, fruiting and fruit quality of sweet pepper cv. California Wonder during autumn 2009 and 2010 seasons. Results indicated that different applied treatments significantly increased all studied growth parameters, namely, number of branches and leaves per plant, leaf area per plant and leaf dry weight. Furthermore, the highest early, marketable and total yields as well as physical characters of sweet pepper fruits were obtained with 100 ppm SA plus chelated 50 ppm zinc followed by 50 ppm SA plus 100 ppm Zn.

Erdal *et al.* (2011) performed a field experiment to study the effects of foliar-applied SA on salt sensitivity, hydrogen peroxide (H_2O_2) generation and activities of antioxidant enzymes like peroxidase (POX) and catalase (CAT) in plant tissues under salt stress. SA treatment significantly increased the fresh and dry weights in both root and shoots of wheat plants under salt stress. SA treatment significantly increased the fresh and dry weights in both root and shoots of wheat plants under salt stress. Similarly, POX and CAT activities were also augmented by SA treatment. In parallel to increasing antioxidative activity, SA treatment decreased H_2O_2content when compared to plants growing under salt stress without SA.

Amin *et al.* (2008) conducted a field experiment for the two successive seasons of 2004-05 and 2005-06 at the experimental station of the National Research Centre at Shalakan, Qalubia Governorate, Egypt to study the response of wheat plants to foliar application of SA, ascorbic acid at (0.0, 100, 200 and 400 mg/L) as well as their interaction on vegetative growth, photosynthetic pigments content, yield and some biochemical constituents of wheat grains. The data indicated that, an enhancement effect of growth characters, yield, total carbohydrate as well as nitrogen, phosphorus and potassium content in wheat grains was obtained by 100 or 200 mg/L of SA. Moreover, the same preceding underwent a reverse pattern of change using the higher concentrations of SA (400 mg/L).

Yordanova and Popova (2007) carried out a field experiment to study the effects of SA and cold on photosynthesis, activities of carboxylating enzymes ribulose-1,5-bisphosphate carboxylase (RuBPC) and phosphoenol pyruvate carboxylase (PEPC) and activities of photo respiratory enzymes glycolate oxidase (GO) and catalase (CAT), and on the activities of antioxidant enzymes superoxide dismutase (SOD), ascorbate peroxidase (APX), glutathione reeducates' (GR) and peroxidase(POX) in winter wheat (*Triticum aestivum,* cv.Dogu-88) leaves. Exposure of wheat plants to a low temperature (3^oC) for 48 h and 72 h resulted in decreased levels of chlorophyll, CO2 assimilation and transpiration rates and increased activity of GO and CAT. Treatment with SA alone for 24 h resulted in a lower rate of photosynthesis, decreased transpiration and stomatal conductance accompanied with enhanced rate of lipid per oxidation and peroxides level. Treatment with

500 mM SA for 24 h before exposure to chilling provided protection on RuBPC activity and chlorophyll content. The activities of GO, CAT, POX, and APX additionally increased in SA-treated plants.

Waseem *et al.*(2006) conducted an experiment to assess whether exogenously applied SA through the rooting medium could mitigate the adverse effects of water stress on plant growth, photosynthesis and nutrient status of two wheat genotypes (S-24 & MH-97). Results showed that different levels of SA applied through the rooting medium increased photosynthetic rate in both cultivars under non-stress conditions but only in S-24 under water stress conditions. Exogenous application of 5 or 10 mg/L SA caused an increase in stomatal conductance, transpiration rate, and sub-stomatal of water stressed plants of cv. S-24 whereas it was true for droughted plants of MH-97 only when 5 mg/L SA applied. Cultivar S-24 was generally higher in N and P contents of shoot and root than that in genotype MH-97 under both normal and water stress conditions. Although, exogenously applied SA through the rooting medium had growth promoting effects under non-stress conditions, it did not mitigate the adverse effects of drought stress on growth of both cultivars, though genotype MH-97 showed some recovery under water stress conditions.

Shakirova *et al.* (2003) reported that wheat seedlings treated with 50 µM SA develop larger ears and increased the level of cell division within the apical meristem of seedling roots causing an increase in plant growth and an elevated wheat productivity. It was found that SA treatment caused accumulation of both abscisic acid (ABA) and indoleacetic acid (IAA) in the wheat seedlings but did not influence cytokinin content. SA treatment reduced the damaging action of salinity on seedling growth and accelerated reparation of the growth processes. SA-treatment diminished changes in phytohormones levels in wheat seedlings under salinity. It prevented any decrease in IAA and cytokinin contents and thus reduced stress-induced inhibition of plant growth. A high ABA level was also maintained in SA-treated wheat seedlings providing the development of antistress reactions, for example, maintenance of proline accumulation. Thus SA's protective action includes the development of antistress programs and acceleration of growth processes recovery after the removal of stress factors.

Gutie´rrez-Coronado *et al. (*1998) showed that SA has a growth-stimulating effect in soybean plants. When soybean plants treated with 10 µM, 100 µM, and up to 100µM SA, shoot and root growth increase 20% and 45% respectively, atseven days after application.

MATERIALS

AND

METHODS

Chapter3

MATERIALS AND METHOD

This chapter deals with the materials and method used in the experiment. It includes a short description of location of the experimental plot, characteristics of soil, climate, crops, treatments, experimental design followed, land preparation, seedling transplanting, intercultural operations, harvesting, data recording, collection and statistical analysis used for the experiment. The details of the experiment are given below.

3.1 Location of the experiment

The research work was carried out at the experimental field of Sher-e-Bangla Agricultural University, Dhaka. The soil of the experimental plots belonged to the Agro Ecological Zone Madhupur Tract (AEZ-28).

3.2 Soil

The farm belongs to the General soil type, **"**Red Brown Terrace Soil**"** under Tejgaon soil Series. The land was above flood level and sufficient sunshine was available during the experimental period. The soil characteristics were clay loam in texture with pH value of 6.3. The morphological, physical and chemical characteristics of initial soil are presented in Tables 1 and 4.

3.3 Climate

The experimental area is under the subtropical climate. Usually the rainfall was heavy during Kharif season and scanty in Rabi season. The atmospheric temperatures increased as the growing period proceeded towards Kharif season. The weather conditions of crop growth period such as monthly mean rainfall (mm), mean temperature (^{O}C), sunshine hours and humidity (%) are presented in Appendix III.

3.4 Planting material

Seed of BRRI hybrid dhan 3 was collected from BRRI, Gazipur. BRRI developed this variety and released in 2009. It is a most popular variety now due to its high yielding potentials and suitable for planted at 15th December-15th January. This variety attains a height of 110cm. The life cycle of this variety is 145 days. Grain yield is 9.21 t/ha and 1000 grain weight is 32-35g.

Table 1. Morphological characteristics of the experimental field

Morphological features	Characteristics
Location	Sher-e-Bangla Agricultural University Farm, Dhaka
AEZ	Madhupur Tract(AEZ 28)
General Soil Type	Red Brown Terrace Soil
Land type	Medium high land
Soil series	Tejgaon
Topography	Fairly leveled
Flood level	Above flood level
Drainage	Well drained

3.5 Seed sprouting:

Healthy seeds were collected by specific gravity method. The selected seeds were soaked for 24 hours and then these were kept in gunny bags. The seed started sprouting after 48 hours and almost all seeds were sprouted after 72 hours.

3.6 Seedbed preparation and seed sowing

Seedbed was prepared on 20 November 2014 for sowing the sprouting seeds and proper care was taken for raising seedlings. The seedbed soil of those locations were cleaning, wetting by proper irrigation and leveling with ladder. Sprouted seeds were sown in the wet soil on 23 November 2014. Weeds were removed and irrigation was given in the seedbed as and when necessary.

3.7 Land preparation

The land was first opened with the tractor drawn disc plough. Ploughed soil was then brought into desirable fine tilth by 4 operations of ploughing and harrowing with country plough and ladder. The stubble and weeds were removed. The first ploughing was done 28 December and the final land preparation was done 30 December, 2014. Experimental land was divided into unit plots following the design of experiment.

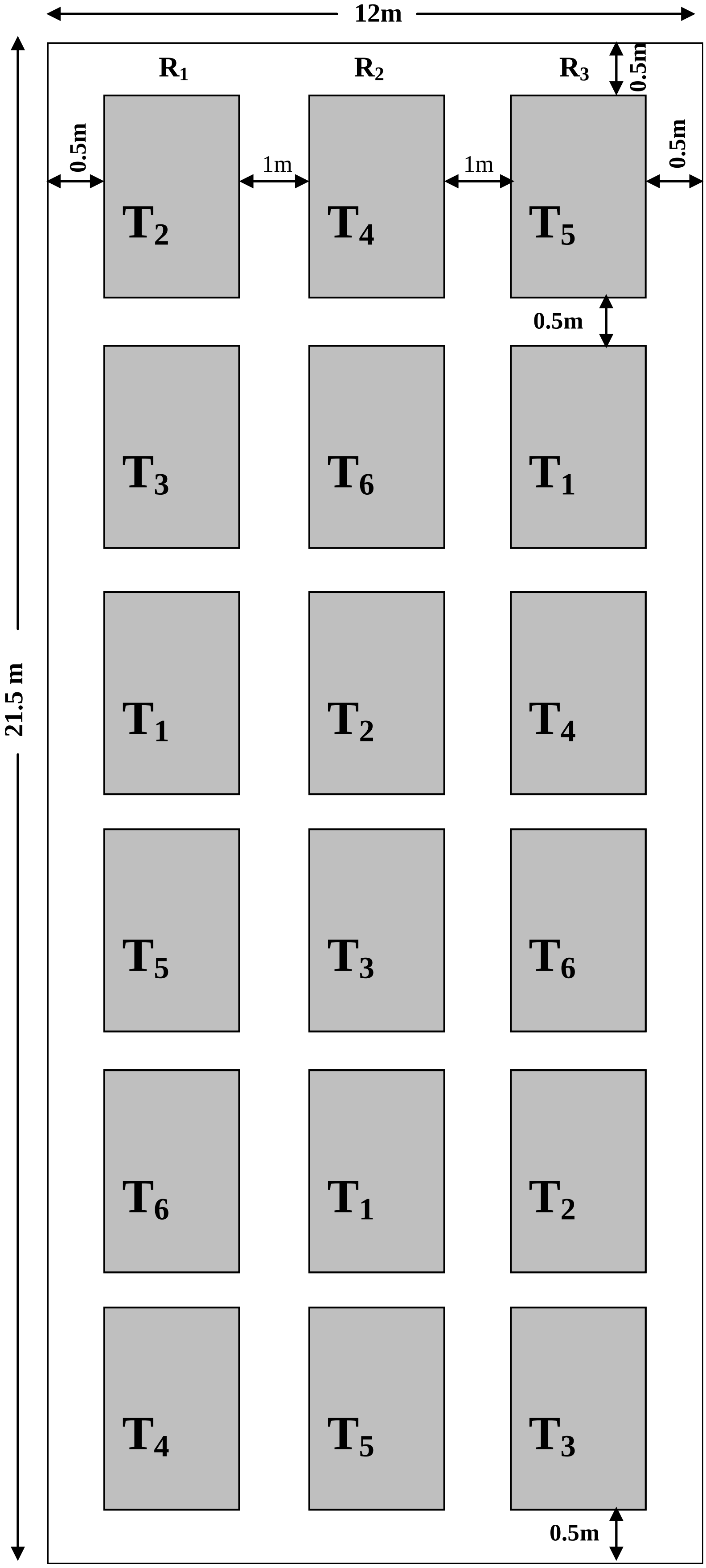
3.8 Treatments of the experiment

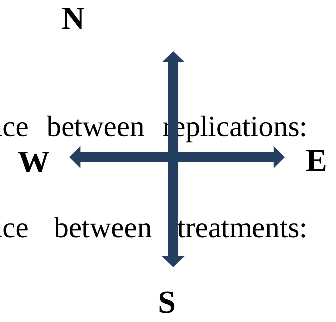
The levels of SA (µM) were as follows:

T_1: 0, T_2: 200, T_3:400, T_4: 600, T_5:800 and T_6: 1000

3.9 Experimental design and layout

The experiment was laid out in a randomized complete block design (factorial). Each treatment was replicated three times. The size of a unit plot was 3 m × 3 m. Total plots in the experimental field were 18. The treatments were randomly distributed to each block. The distance between two adjacent replications (block) was 1m and row-to-row distance was 0.5 m. The inter block and inter row spaces were used as footpath and irrigation or drainage channel.





Distance between replications: 1m

Distance between treatments: 0.5m

Total length of the field:
(6×3m) + (7×0.5m) = 21.5m

Total width of the field:
(3×3m)+(0.5m+1m+1m+0.5m= 12m

Total area of the field:
L×B = 21.5m × 12m = 258m^2

Replications:

R_1: Replication 1
R_2: Replication 2
R_3: Replication 3

Treatments:

T1: 0 µM Salicylic acid

T2: 200 µM Salicylic acid

T3: 400 µM Salicylic acid

T4: 600 µM Salicylic acid

T5: 800 µM Salicylic acid

T6: 1000 µM Salicylic acid

Fig.1. Layout of experimental plot

3.10 Fertilizer application

Every treatment received N, P and K as basal dose. The rate and source of nutrients used in the study is given in Table2. All the fertilizers except N were added to the soil during final land preparation on 30 December,

2014. Urea was splitted based on Adhunik Dhaner Chash, BRRI (2015).

Table2. Name of the element, rate and name of the fertilizer used for the experiment

Name of the element	Rate (kg/ha)	Source of nutrient
N	120	Urea
P	20	Triple Super Phosphate(TSP)
K	60	Muriate of Potash (MoP)
S	16	Gypsum
Zn	2	Zinc sulphate

Ref. Fertilizer Recommendation Guide, 2012.

3.11 Transplanting of seedling

Thirty-seven-day old seedlings were uprooted carefully from the seedbed and transplanted in the experimental plots on 30 December, 2014, using 20 x 20 cm spacing with one seedling per hill.

3.12 Intercultural operations

Intercultural operations were done for ensuring and maintaining the normal growth of the crop. The detailed intercultural operations were recorded in the Table3.

3.12.1 Irrigation

After transplanting 5-6 cm water was maintained in each plot through irrigation up to hard dough stage.

3.12.2 Weeding

The crop was infested with some common weeds, which were controlled by uprooting and removed them at 20, 30 and 45 days after transplanting (DAT).

Table 3 . Dates of different operations done during the field study

Operations	Working Dates
First ploughing of the field	28 December 2014
final land preparation	30 December 2014
Application of fertilizers (1/4thUrea, TSP, MP, Gypsum,	30 December 2014

$ZnSO_4$ as basal)	
Transplanting of seedlings	30 December 2014
Intercultural Operations	**Working Dates**
2nd split application of urea and weeding	20 January 2015
3rd split application of urea and 2nd weeding	8 February 2015
First step of SA application	16 February 2015
Insecticide application	2 March 2015
Second step of SA application Third step of SA application	23 March 2015 1 April 2015
Harvesting and threshing	2 May 2015

3.12.3 Protection against insect and pest

There were some incidence in insects specially rice stem borer, grasshopper, rice bug etc. which were controlled by spraying Diazinon 50EC.

3.13 Preparation and application of Salicylic acid:

The mixture of 13.812 g SA in 100 mL ethanol is called 1M Stock solution of SA and 0.4 mL SA solution was taken from stock solution in 2 L spray bottle for making 200 µM SA solution. Similarly 1mL and 2mL SA solution were taken from stock solution in 2 L spray bottle for making 500 µM and 1000 µM SA solution respectively. Foliar application of SA was done in rice field in three times. First step was applied on 49DAT (tillering stage). 2nd applications on 84DAT (panicle initiation stage) and 3rd applications on 92DAT (flowering stage).

3.14 Crop sampling and data collection

The crop sampling was done at the time of harvest. Harvesting date was 02/5/2015. At each harvest, ten plants were selected randomly from each plot. The selected plants of each plot were cut carefully at the soil surface level. The plant heights, panicle length, number of grain/panicle, 1000 grain weight and yield were recorded separately.

3.15 Harvest and Threshing

Harvesting was done when 90% of the crops became brown in color. The matured crop were cut and collected manually from a pre demarcated area of 5 m^2 at the centre of each plot. The harvested crops were threshed, cleaned and processed. Grain yield and straw was recorded plot wise and moisture of straw was calculated on oven dry basis. Grain yield was adjusted at 14% moisture level.

3.16 Data collection

Ten hills were selected randomly from each plot prior to harvest for recording data on crop parameters and the yield of grain straw were taken plot wise.

The following parameters were recorded at harvest:

1) Fresh weight of 10 hills at maximum tillering stage
2) Oven dry weight of 10 hills at maximum tillering stage
3) Plant height (cm) from soil base to tip of panicle
4) Panicle length (cm)
5) Number of affected panicle/plot
6) Number of effective tillers/hill
7) Number of filled spikelet or grain/panicle
8) Unfilled spikelet or chita/panicle
9) 1000 grain weight
10) Grain yield
11) Straw yield (t/ha)
12) Biological yield (t/ha)
13) Soil sample collection from each plot (post-harvest)
14) Recording insect pest infestation data

3.17 Procedure of data collection:

3.17.1 Fresh weight of ten hills at maximum tillering stage

Ten hills from each plot were collected at maximum tillering stage and then weighted by using a digital electric balance.

3.17.2 Oven dry weight of 10 hills at maximum tillering stage

Ten hills from each plot were collected at maximum tillering stage and then sun dried. The sun dried hills again dried in oven and weighted by using a digital electric balance.

3.17.3 Canopy height

The height of ten plants was measured with a meter scale from the ground level to tip of the plants and the mean height was expressed in cm.

3.17.4 Panicle length

Panicle length was measured from the basal node of rachis to the tip of panicle.

3.17.5 Number of affected panicle/plot

Number of affected panicle per plot was counted.

3.17.6 Number of effective tillers/hill

Total number of effective tillers/hill from each plot was counted at the time of harvest.

3.17.7 Number of filled spikelet or grain/panicle

Total grain numbers were counted from total panicle that was obtained from pre-selected ten plants. After that it was averaged and expressed as number of grain per panicle.

3.17.8 Number of unfilled spikelet or chita/ panicle

Unfilled grain per panicle were counted from ten hills and then averaged.

3.17.9 Weight of 1000 grain

One thousand cleaned dried grains were counted randomly from each harvest sample and weighted by using a digital electric balance and mean weight was expressed in gram.

3.17.10 Grain yield (t/ha)

Weight of grain of the demarcated area ($5m^2$) of each plot was taken and then converted to the yield in t/ha.

3.17.11 Straw yield (t/ha)

Straw obtained from each plot were sun dried and weighted carefully. The dry weight was taken carefully. The dry weighted straw of central $5m^2$was taken and straw yield/plot and finally converted to t/ha.

3.17.12 Biological yield (t/ha)

Biological yield was calculated from addition of grain yield and straw yield.

3.17.13 Harvest Index (%)

The harvest index (HI) was calculated by dividing the actual grain yield by the biological yield of the crop. It was expressed as percentage.

$$\text{Harvest Index} = \frac{\text{Grain yield(t/ ha)}}{\text{Biological yield(t/ ha)}} \times 100$$

3.17.14 Recording of insect-pest infestation data

Major insect-pest infestations were observed carefully and whiteheads symptoms were observed at the heading stage. All the whitish panicles were collected plot-wise and recorded them. Finally average the affected panicle numbers according to the treatments.

3.18 Analysis of data

The data collected on different parameters were statistically analyzed to obtain the level of significance using the Statistic10software. 5% level of significance was used to compare the mean difference among the treatments.

3.19 Collection and preparation of initial soil sample

The initial soil samples were collected before land preparation from a 0-15 cm soil depth. The sample was drawn by means of an auger from different locations covering the whole experimental plot mixed thoroughly to make a composite sample. After collection of soil samples, the plant roots, leaves etc. were picked up and removed. Then the samples were dried and sieved through a 10-mesh sieve and stored in a clean plastic container for physical and chemical analysis.

3.20 Chemical analysis of soil sample

Soil sample was analyzed for both physical and chemical properties in the laboratory of Soil Science Department, Sher-e-Bangla Agricultural University, Dhaka. The properties studied included soil texture, p^H, organic matter, total N, available P, exchangeable K, and available S (Table 4). The physical and chemical properties of postharvest soil have been presented in Appendix II. The soil was analyzed by standard methods:

Table4. Physical and chemical properties of the initial soil sample

Characteristics	Value
Particle size analysis	
% Sand	30
% Silt	40
% Clay	30
Textural class	Clay loam
Consistency	Granular and friable when dry
pH	6.3
Bulk Density (g/cc)	1.45

Particle Density (g/cc)	2.53
Organic carbon (%)	0.61
Organic matter (%)	1.05
Total N (%)	0.06
Available P (ppm)	20
Exchangeable K (meq/100g soil)	0.12
Available S (ppm)	22

3.21 Particle size analysis

Particle size analysis of soil was done by Hydrometer Method and then textural class was determined by plotting the values for %sand, % silt and % clay to the "Marshall's Textural Triangular Coordinate" according to the USDA system.

3.22 Soil P^H

Soil p^H was measured with the help of a Glass electrode p^H meter using soil and water at the ratio of 1:2.5 as described by Jackson (1962).

3.23 Organic C

Organic carbon in soil was determined by Walkley and Black (1934) Wet Oxidation Method. To obtain the organic matter content, the amount of organic carbon was multiplied by the Van Bemmelen factor, 1.73. The result was expressed as percentage.

3.24 Total nitrogen

Total N content of soil were determined followed by the Micro Kjeldahl method. One gram of oven dry ground soil sample was taken into micro kjeldahl flask to which 1.1 gm catalyst mixture (K_2SO_4: $CuSO_4$. $5H_2O$: Se in the ratio of 100:10:1), and 6 ml H_2SO_4 were added. The flasks were swirled and heated 200^0C and added 3 ml H_2O_2 and then heating at 360^0C was continued until the digest was clear and colorless. After cooling, the content was taken into 100 ml volumetric flask and the volume was made up to the mark with distilled water. A reagent blank was prepared in a similar manner. These digests were used for nitrogen determination. Then 20 ml digest solution was transferred into the distillation flask, Then 10 ml of H_3BO_3 indicator solution was taken into a 250 ml conical flask which is marked to indicate a volume of 50 ml and placed the flask under the condenser outlet of the distillation apparatus so that the delivery end dipped in the acid. Add sufficient amount of 10N-NaOH solutions in the container connecting with distillation apparatus. Water runs through the condenser of distillation apparatus was checked. Operating switch of the distillation apparatus collected the distillate. The conical flask was removed by washing the delivery outlet of the distillation apparatus with distilled water. Finally the distillates were titrated with standard 0.01 N H_2SO_4 until the color changes from green to pink. The amount of N was calculated using the following formula:

$$\% N = (T-B) \times N \times 0.014 \times 100/S$$

Where,T = Sample titration (ml) value of standard H_2SO_4

B = Blank titration (ml) value of standard H_2SO_4

N = Strength of H_2SO_4,S = Sample weight in gram

3.25 Available phosphorus

Available P was extracted from the soil with 0.5 M $NaHCO_3$ solutions, pH8.5 (Olsen *et al.*, 1954). Phosphorus in the extract was then determined by developing blue color with reduction of phosphomolybdate complex and the color intensity were measured colorimetrically at 660 nm wavelength and readings were calibrated with the standard P curve.

3.26 Exchangeable potassium

Exchangeable K was determined by 1N NH_4OAc (pH 7) extraction methods and by using flame photometer and calibrated with a standard curve.

RESULTS
AND
DISCUSSIONS

Chapter4

RESULTS AND DISCUSSION

The results of the experiment conducted under field conditions are presented in several tables and figures. The experiment was conducted to study the effect of different levels of salicylic acid on the performance of BRRI hybrid dhan3. The results are presented and discussed under following parameters.

4.1 Role of Salicylic acid (SA) at maximum tillering stage

4.1.1 On fresh biomass production

A significant variation was observed in the biomass production of BRRI hybrid dhan3 due to the foliar application of SA at the maximum tillering stage. Fresh weight production was increased with increasing the level of SA up to the 400 µM and then decreased due to the higher doses of SA up to the 1000 µM. The maximum fresh weight (15.0 t/ha) was produced in T_3 treatment and minimum (12.8 t/ha) was in the T_1 treatment (Table 5). However, the reduction of the biomass production was not less the control treatment, T_1 due to the higher dose of SA in the treatment T_6 (12.8 t/ha) and it was statistically similar with the control treatment. Farooq *et al.,* (2009) found that seedling fresh weight increased by the application of salicylic acid.

4.1.2 On dry matter production

Dry mass production of BRRI hybrid dhan3 differed significantly among the treatment combinations (Table 5). The highest oven dry weight (2.7 t/ha) was found in the T_3 treatment having foliar spray of SA at 400 µM and the lowest oven dry weight (2.0 t/ha) was found in the treatment, T_5 having 800 µM of SA, which was statistically similar with the control treatment T_1 (2.1 t/ha) having no SA. This results indicate that foliar application of SA at lower dose has positive effect on the dry mass production of BRRI hybrid dhan3 and the higher doses of SA up to 1000 µM have no positive effect on the dry mass production (Table 5).Dry mass production increased due to application of SA (Usharani *et al*.,2014).

Table 5. Effect of different treatments on fresh and dry matter production of BRRI hybrid dhan3 at maximum tillering stage, Boro 2014-15, SAU,Dhaka

Treatments	Fresh biomass (t/ha)	Dry matter (t/ha)
T_1=0 µM Salicylic Acid	12.8 c	2.1 b
T_2=200 µM Salicylic Acid	13.7 b	2.2 b
T_3=400 µM Salicylic Acid	15.0 a	2.7 a
T_4=600 µM Salicylic Acid	14.2 b	2.2 b
T_5=800 µM Salicylic Acid	12.9 c	2.0 b
T_6=1000 µM Salicylic Acid	12.8 c	2.1 b
$LSD_{0.05}$	0.55	0.30
SE (±)	0.25	0.11
Level of significance	**	*
CV (%)	2.22	7.28

** = Significant at 1% level of probability, * = Significant at 5% level of probability

4.1.3 Canopy height

The canopy height of BRRI hybrid dhan3 was varied significantly due to the different treatment combinations. The tallest plant (87.0 cm) was found in the T_3 treatment having 400 µM of SA and it was statistically identical with the treatments T_2, T_4 and T_5. The shortest plant (82.4 cm) was found in the treatment T_6 due to the application of 1000 µM of SA. There were no significant differences between the control treatment T_1 and T_6 (Table 6). These results indicate that lower dose of SA (200-400 µM) has a positive role on plant height of BRRI hybrid dhan3 and higher doses up to (600-1000 µM) have negative effect on the canopy height at harvesting stage. Singh *et al*., (2015) and Usharani *et al*., (2014) found that foliar application of SA increases plant height of rice significantly.

4.1.4 Number of effective tillers/hill

The application of different levels of SA had a significant effect on number of effective tillers per hill. Number of effective tillers increased with the increases of SA levels. The highest number of effective tillers per hill (14) was found in the treatment T_6 due to the application of 1000 µM SA which was statistically similar with the treatments T_3, T_4 and T_5 (Table 6). The lowest number of effective tillers per hill (12) was found in the control, T_1 treatment. These results might be due to the optimum use of irrigation water because foliar application of SA reduces the transpirational water losses and increases the total chlorophyll levels in the leaves. Singh *et al.* (2015) found that foliar application of SA significantly increases number of effective tillers/hill of rice. Sardoei*et al.*,(2014) reported that the exogenous spray of SA had significant effect on number of tillers.

4.1.5 1000 grain weight (g)

The 1000 grain weight of BRRI hybrid dhan3 showed significant difference due to the foliar application of different levels of SA. The maximum 1000 grain weight (34.8g) was found in the treatment T_6 having 1000 µM SA which was statistically similar to the treatment T_3, T_4 and T_5. The lowest 1000 grain weight (29.3g) was found in the control treatment T_1 which was statistically similar to the treatment T_2, T_3, T_4 and T_5 (Table 6). Ibrahim *et al.,* (2014) showed 1000 grain weight increased significantly by the application of salicylic acid.

Table 6. Effect of different treatments on plant height, effective tillers/hill and 1000 grain weight of BRRI hybrid dhan3 at harvest, Boro 2014-15, SAU, Dhaka

Treatments	Plant height (cm)	Effective tillers/hill	1000 grain weight (g)
T1=0 µM Salicylic Acid	82.8 b	12 b	29.3 b

T2=200 µM Salicylic Acid	84.0 ab	12 ab	30.0 ab
T3=400 µM Salicylic Acid	87.0 a	13 ab	32.0 ab
T4=600 µM Salicylic Acid	86.2 a	13 a	32.7 ab
T5=800 µM Salicylic Acid	83.9 ab	13 a	33.7 ab
T6=1000 µM Salicylic Acid	82.4 b	14 a	34.8 a
$LSD_{0.05}$	3.15	1.35	4.93
SE (±)	1.00	0.61	2.21
Level of significance	*	*	*
CV (%)	2.05	5.79	8.45

** = Significant at 1% level of probability, * = Significant at 5% level of probability

4.1.6 Panicle length (cm)

The panicle length of BRRI hybrid dhan3 was significantly affected by the different levels of SA. The highest panicle length (25.5cm) was found in the T_3 treatment having foliar application of 400 µM SA and it was statistically similar with control treatment, T_1 and T_6 treatment (Table 7). The lowest panicle length (23.6 cm) was found in the T_2 treatment having foliar application of 200 µM SA and it was also statistically similar with T_1, T_4, T_5 and T_6 treatments.

4.1.7 Total spikelet /panicle

The influence of SA on the number of total spikelet/panicle was significantly varied among the treatments. Number of spikelet/panicle was significantly influenced by the foliar application of SA at different doses. The highest number of spikelet was found in T_6 treatment which was statistically similar with T_1 and T_5 and the lowest was found in T_2-T_4 treatments.

4.1.8 Filled spikelet/ panicle

The foliar application of different levels of SA had a significant effect on filled spikelet per panicle. The highest number of grain per panicle (156) was found in the T_6 treatment having foliar application of 1000 µM SA and it was statistically similar with the treatments T_4 and T_5 (Table 7) which indicate that higher doses of SA (600-1000 µM) have a positive role on the spikelet fertility of BRRI hybrid dhan3. The lowest number of filled grain per panicle (141) was found in the T_1 treatment and it was statistically similar with T_2 and T_3 treatments (Table 7). This result suggests that foliar application of SA could help to increase the grain yield of BRRI hybrid dhan3. Singh *et al.*, (2015) and Usharani *et al*., (2014) showed that filled grain/panicle increased significantly by the application of salicylic acid.

Table 7. Effect of foliar application of SA on panicle length and number of spikelet /panicle of BRRI hybrid dhan3at harvest, Boro 2014-15, SAU, Dhaka

Treatments	Panicle length (cm)	Total Spikelet/ panicle	Filled spikelet/ panicle	Filled spikelet increased over control T_1 (%)
T1=0 µM Salicylic Acid	24.8 ab	166 a	141 b	-
T2=200 µM Salicylic Acid	23.6 b	160 b	143 b	1.4
T3=400 µM Salicylic Acid	25.5 a	158 b	146 b	3.4
T4=600 µM Salicylic Acid	24.1 b	161 b	151 a	6.6
T5=800 µM Salicylic Acid	24.0 b	166 a	153 a	7.8
T6=1000 µM Salicylic Acid	24.6 ab	167 a	156 a	9.6
$LSD_{0.05}$	1.27	7.85	9.26	
SE (±)	0.403	2.49	2.94	
Level of significance	NS	*	**	
CV (%)	2.86	2.68	3.46	

** = Significant at 1% level of probability, * = Significant at 5% level of probability, NS = Not significant

4.1.9 Unfilled spikelet/panicle and sterility (%)

The application of different levels of SA had a significant effect on the unfilled spikelet per panicle. The lowest number of unfilled spikelet per panicle (11) was found in application of 1000 µM SA under the treatment T_6 which was similar with T_2, T_4 and T_5. The highest number of unfilled spikelet per panicle (25) was found in the control treatment T_1 which was similar with T_2 and T_3 (Table 8). Sterility (%) also decreased with increases of SA level. These results showed that number of unfilled spikelet per panicle was decreased with increases of level of SA as foliar application. Mohammed, R.A. (2011) reported that number of unfilled spikelet per panicle was decreased due to foliar application of SA.

Table 8. Effect of foliar application of SA on spikelet sterility (%) of BRRI hybrid dhan3 at harvest, Boro 2014-15, SAU, Dhaka

Treatments	Unfilled spikelet/ panicle	Spikelet sterility (%)
T1=0 µM Salicylic Acid	25 a	15.1
T2=200 µM Salicylic Acid	17 abc	10.6
T3=400 µM Salicylic Acid	12 ab	7.6
T4=600 µM Salicylic Acid	11 bc	6.8
T5=800 µM Salicylic Acid	12 bc	7.2
T6=1000 µM Salicylic Acid	11 c	6.7
$LSD_{0.05}$	1.48	
SE (±)	0.472	
Level of significance	*	
CV (%)	6.93	

* = Significant at 5% level of probability

4.1.10 Number of stem borer affected panicle/plot

The application of different levels of SA had a significant effect on number of stem borer affected plants. The lowest number of affected panicle per plot (3) was found in the treatment T_6which was similar with $T_5$and this might be due to the application of higher SA (1000 µM). The highest number of affected panicle per plot (12) was found in the control treatment T_1which was similar with T_2(Table 9). Number of stem borer affected plants was decreased gradually with increases SA levels up to 1000 µM. This result suggests that foliar application of SA could be effective on rice stem borer infestation management. Insect infestation reduced significantly due to application of SA (Singh *et al.* 2015).

Table 9. Effect of treatments on number of affected panicle/plot of BRRI hybrid dhan3, Boro 2014-15, SAU, Dhaka

Treatments	Number of affected panicle/plot
T1=0 µM Salicylic Acid	12 a
T2=200 µM Salicylic Acid	10 a
T3=400 µM Salicylic Acid	7 b
T4=600 µM Salicylic Acid	8 b
T5=800 µM Salicylic Acid	4 c
T6=1000 µM Salicylic Acid	3 c
$LSD_{0.05}$	1.68
SE (±)	0.534
Level of significance	**
CV (%)	12.52

** = Significant at 1% level of probability

4.1.11 Grain yield

Grain yield increased with increases the level of SA as a foliar application at different stages of BRRI hybrid dhan3. The maximum grain yield was found in the treatment T_6 having 1000 µM SA and it was statistically identical with the treatment T_5 having 800 µM SA. On the other hand, minimum grain yield was found in the control treatment T_1 which was similar to the treatments T_2 and T_3 having 200 µM and 400 µM SA, respectively. Among the treatment combinations, T_4 to T_6 treatments significantly increased the grain yield. Compare to the control treatment, grain yield was increased 5.6% - 31.4% due to the foliar application of SA in the BRRI hybrid dhan3 (Table 10). Although,potential grain yield of BRRI hybrid dhan3 was recorded 9.00 t/ha at the research satiation, here it was found 7.01 t/ha. However, foliar application of salicylic acid could be helpful to recover the yield gap, increasing the grain yield and reduction of insect-pest infestation. Sharafizad*et al.*, (2012) showed that dosage of SA significantly affected total grain yield.

4.1.12 Straw yield (t /ha)

The effect of different levels of SA on straw yield of BRRI hybrid dhan3 was highly significant. The straw yield of BRRI hybrid dhan3 increased significantly due to foliar application of SA at 1000 μM under the treatment T_6**.** The maximum straw yield (9.22 t/ha) was observed from the T_6 treatment having 1000 μM SA which was statistically similar with T_5 treatment (Table 10). The minimum straw yield (7.23 t/ ha) was obtained from the control treatment T_1 (having no SA) which was statistically identical with the treatments T_2, T_3, and T_4. Compare to the control treatment straw yield was increased 7.8% - 27.5% due to the foliar application of SA in the BRRI hybrid dhan3 (Table 10). Usharani*et al*., (2014) showed highest straw yield was achieved by the application of salicylic acid.

4.1.13 Biological Yield

Significant response was observed in the biological yield of BRRI hybrid dhan3 due to foliar application of different level of SA. The biological yield was varied from 14.25 - 18.56 t /ha. The highest biological yield (18.56 t/ha) was obtained in the T_6 treatment which was statistically similar with the treatment T_5. On the other hand, lowest biological yield (14.25 t/ha) was obtained from the T_1 treatment and it was statistically identical with the treatments T_2, T_3, and T_4(Table 10).

4.1.14 Harvest Index

Significant response was not observed in the harvest index due to the foliar application of different levels of SA on BRRI hybrid dhan3. From the results, it was found that the highest harvest index (50.6%) was obtained from the treatment T_6 and the lowest index (48.7%) was obtained in the T_2 treatment whereas, treatment T_1 having the harvest index 49.2% (Table 10)

Table 10.Effect of treatments on grain, straw, biological yield and harvest index of BRRI hybrid dhan3, Boro 2014-15, SAU, Dhakarice

Treatments	Grain yield (t/ha)	Straw (t/ha)	Biological yield (t/ha)	Harvest index (%)
T1=0 μM Salicylic Acid	7.01 d	7.23 c	14.24 b	49.2
T2=200 μM Salicylic Acid	7.40 cd	7.80 bc	15.20 b	48.7
T3=400 μM Salicylic Acid	7.75 bcd	7.72 bc	15.47 b	50.1
T4=600 μM Salicylic Acid	8.21 bc	8.01 bc	16.22 b	50.6

T5=800 µM Salicylic Acid	8.65 ab	8.51 ab	17.16 a	50.4
T6=1000 µM Salicylic Acid	9.21 a	9.22 a	18.43 a	50.0
$LSD_{0.05}$	1.02	1.09	1.44	4.16
SE (±)	0.324	0.345	0.457	1.32
Level of significance	**	*	**	NS
CV (%)	6.94	7.40	4.90	4.58

** = Significant at 1% level of probability, * = Significant at 5% level of probability, NS = Not significant

Table 11.Grain yield and straw increase over control of BRRI hybrid dhan3, Boro 2014-15, SAU, Dhakarice

Treatments	Grain yield (t/ha)	% Increase over control	Straw yield (t/ha)	% Increase over control
T1=0 µM Salicylic Acid	7.01 d		7.23 c	-
T2=200 µM Salicylic Acid	7.40 cd	5.6	7.80 bc	7.8
T3=400 µM Salicylic Acid	7.75 bcd	10.6	7.72 bc	6.7
T4=600 µM Salicylic Acid	8.21 bc	17.1	8.01 bc	10.8
T5=800 µM Salicylic Acid	8.65 ab	23.4	8.51 ab	17.7
T6=1000 µM Salicylic Acid	9.21 a	31.4	9.22 a	27.5
$LSD_{0.05}$	1.02	-	1.09	-
SE (±)	0.324	-	0.345	-
Level of significance	**	-	*	-
CV (%)	6.94	-	7.40	-

** = Significant at 1% level of probability, * = Significant at 5% level of probability, NS = Not significant

SUMMARY
AND
CONCLUSION

Chapter 5

SUMMARY AND CONCLUSION

The experiment was conducted in the Farm of Sher-e-Bangla Agricultural University, Dhaka, Bangladesh during the period of November 2014 - April 2015 to evaluate the effect of different levels of SA (0, 200, 400,600, 800and 1000µM) on grain yield and yield attributes of BRRI hybriddhan3. The soil of the experimental field belongs to Madhupur tract (AEZ-28) representing Tejgaon soil series.

The experiment was laid out in a randomized complete block design with three replications. There were 18 unit plots and the size of the plot was 3m × 3m. There were 6 treatments combination. BRRI hybrid dhan3 was sown as test crop. Data on different growth and yield parameters were recorded and **a**nalyzed statistically. Foliar application of SA was done in rice field in three times. First step was applied on 49DAT (tillering stage). 2ndapplications on 84DAT (panicle initiation stage) and 3rdapplications on 92DAT (flowering stage). Weeding, irrigation and pest managements were done if and when necessary. Ten hills at maximum tillering stage from each plot were selected randomly for taking data on fresh biomass production, dry matter production. Plant height, panicle length, number of affected panicle/plot, number of effective tillers/hill, number of filled spikelet/panicle, unfilled spikelet/panicle, 1000 grain weight, grain yield, straw yield, biological yield and harvest index were taken.

The maximum fresh weight (15.0 t/ha) was production in T_3 treatment and minimum (12.8 t/ha) was in the T_1 treatment at maximum tillering stage. The highest oven dry weight (2.7 t/ha) was found in the T_3 treatment having foliar spray of SA at 400 µM and the lowest oven dry weight (2.0 t/ha) was found in the treatment, T_5 having 800 µM of SA, which was statistically similar with the control treatment T_1 (2.1 t/ha) having no SA. The tallest plant

(87.0 cm) was found in the T_3 treatment having 400 µM of SA and it was statistically identical with the treatments T_2, T_4 and T_5. The shortest plant (82.4 cm) was found in the treatment T_6. The lowest number of affected panicle per plot (3) was found in the treatment T_6 which was similar with $T_5$and the highest number of affected panicle per plot (12) was found in the control treatment T_1which was similar with T_2. The highest number of effective tillers per hill (14) was found in the treatment T_6 due to the application of 1000 µM SA which was statistically similar with the treatments T_3, T_4 and T_5. The highest number of spikelet (166/panicle) was found in T_6 treatment which was statistically similar with T_1 and T_5 and the lowest was found in T_2-T_4 treatments. The highest number of grain per panicle (156) was found in the T_6 treatment having foliar application of 1000 µM SA and it was statistically similar with the treatments T_4 and T_5. Similarly, the lowest number of unfilled spikelet per panicle (11) was found with the same treatment. The maximum 1000 grain weight (34.8g) was found in the treatment T_6 having 1000 µM SA which was statistically similar to the treatment T_3, T_4 and T_5.The maximum grain yield (9.21 t/ha) was found in the treatment T_6 having 1000 µM SA and it was statistically identical with the treatment T_5 having 800 µM SA where the control treatment T_1 (7.01 t/ha) was the lowest and the highest straw yield was (9.22 t/ha) in T_6 treatment where the control treatment T_1 was (7.23 t/ha).

The present study was conducted to improve our understanding of rice responses to SA application. Our results indicated beneficial effects of SA application. Application of SA have a profound effect of effective tillers/hills, 1000 grain weight, filled grain/panicle, unfilled grain/panicle, number of affected panicle/plot, grain yield, straw yield & biological yield. Decreased respiration rates and increased membrane integrity as a result of SA application might have increased the amount of photo synthates transported to the grains, thereby increasing the number of filled grains per panicle, hence increased spikelet fertility.

In conclusion, yield, the final manifestation of all the physiological processes, increased as a result of SA application under non-stress conditions. The overall results of the present study demonstrated that rice may be grown successfully for obtaining maximum yield with the application of SA @ 1000µM as foliar application. However, before making conclusion concerning the appropriate dose of salicylic acid, the study needs further investigation in other Agro Ecological Zones (AEZs) of Bangladesh for country-wide recommendation which will be useful.

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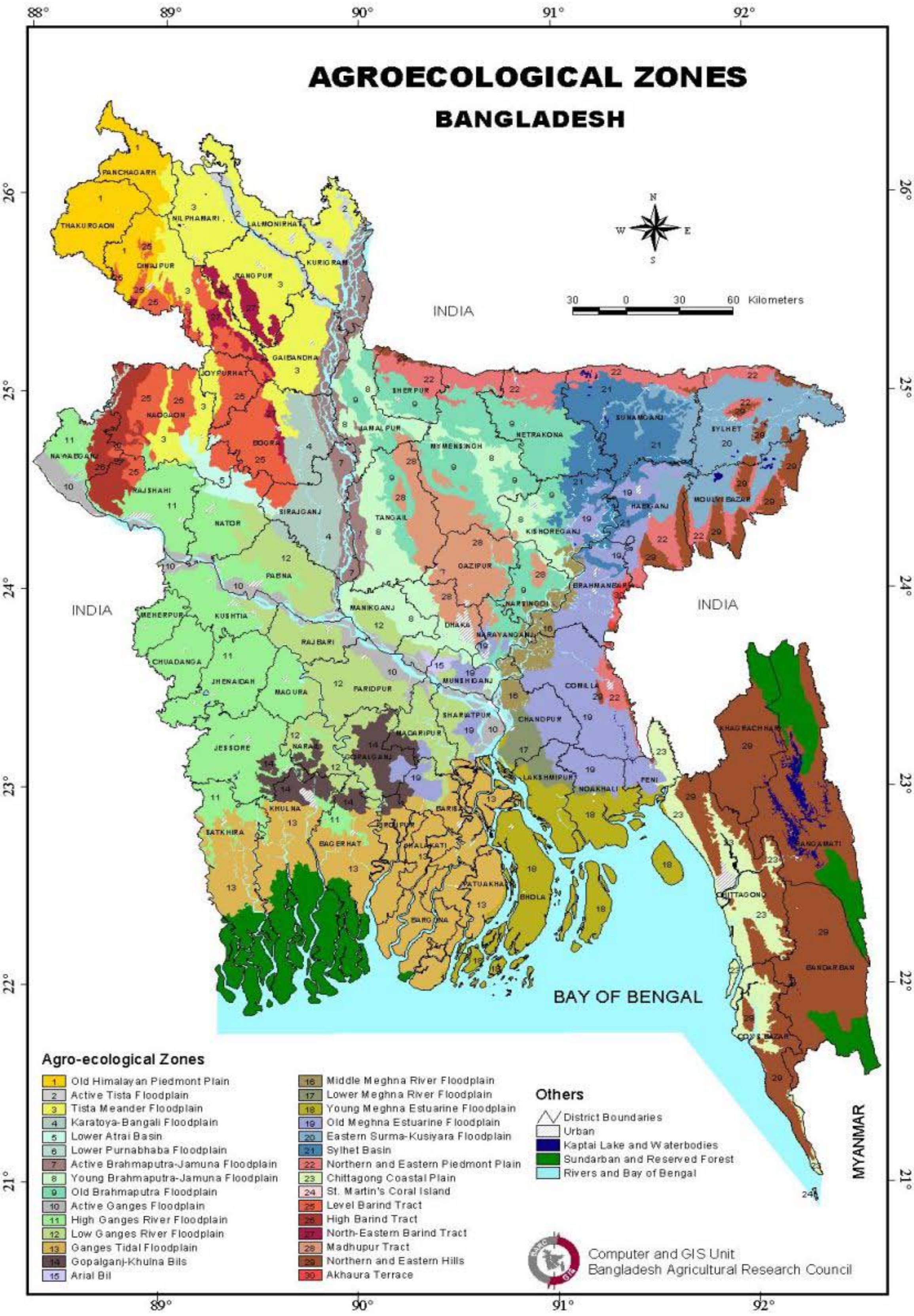
APPENDICES

APPENDICES

Appendix I. Agro-Ecological Zones of Bangladesh



Appendix II. Chemical properties of post-harvest soil

Treatments	% Organic Carbon	% Organic matter	pH
Initial soil	0.61	1.05	6.3
T_1=0 µM Salicylic Acid	0.62	1.07	6.4
T_2=200 µM Salicylic Acid	0.68	1.08	6.4
T_3=400 µM Salicylic Acid	0.69	1.09	6.5
T_4=600 µM Salicylic Acid	0.66	1.05	6.4
T_5=800 µM Salicylic Acid	0.71	1.03	6.4
T_6=1000 µM Salicylic Acid	0.65	1.02	6.5
$LSD_{0.05}$	0.057	0.058	0.645
SE (±)	0.021	0.022	0.205
Level of significance	NS	NS	NS
CV (%)	5.51	3.34	5.27

** = Significant at 1% level of probability, * = Significant at 5% level of probability, NS = Not significant

Appendix III. Monthly record of air temperature, relative humidity, rainfall and sunshine hour of the experimental site during the period from November 2014 to March 2015

Month	*Air temperature (ºc)		*Relative humidity (%)	Total Rainfall (mm)	*Sunshine (hr)
	Maximum	Minimum			
November, 2014	25.8	16.0	78	00	6.8
December, 2014	22.4	13.5	74	00	6.3
January, 2015	24.5	12.4	68	00	5.7
February, 2015	27.1	16.7	67	30	6.7
March, 2015	28.1	19.5	68	00	6.8

* Monthly average,

* **Source**: Bangladesh Meteorological Department (Climate & weather division) Agargoan, Dhaka – 1212