EFFECT OF MODIFIED CHITOSAN ON GROWTH AND YIELD OF RICE (BRRI dhan62)

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DECEMBER, 2015

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A thesis submitted to the faculty of agriculture, Sher-e-Bangla Agricultural University, Dhaka, in partial fulfillment of the requirements for the degree of

MASTER OF SCIENCE (MS)
IN
SOIL SCIENCE
SEMESTER: JULY-DECEMBER, 2015

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CERTIFICATE

This is to certify that thesis entitled, "EFFECT OF MODIFIED CHITOSAN ON GROTH AND YIELD OF RICE (BRRI dhan62)" submitted to the Faculty of Agriculture, Sher-e-Bangla Agricultural University Dhaka, in partial fulfillment of the requirements for the degree of MASTER OF SCIENCE in SOIL SCIENCE, embodies the result of a piece of bona fide research work carried out by MOSTARAK HOSSAIN MUNSHI, Registration No. 09-03547 under my supervision and guidance. No part of the thesis has been submitted 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 been duly acknowledged.

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ACKNOWLEDGEMENTS

The author wishes to acknowledge the immeasurable grace and profound kindness of the "Almighty Allah" the Most Gracious and The Supreme Rule of the universe for giving mental peace, health and strength to submit the thesis for the degree of Master of Science (MS) in Soil Science.

*The author would like to extend his heart-squeezed gratitude, deepest appreciation, best regards and indebtedness to his honorable teacher and research Supervisor **Associate Professor Dr. Mohammad Issak,** Department of Soil Science, Sher-e-Bangla Agricultural University, Sher-e-Bangla Nagar Dhaka, for his untiring guidance, scholastic supervision, valuable advice, innovative suggestions, constant encouragement, helpful comment, affectionate feeling and inspiration in all phases of conducting the research work and preparation of the thesis.*

*The author would like to express his sincere appreciation, heartfelt gratitude and immense indebtedness to his research Co-supervisor. He also expresses his respect to the honorable Co-supervisor, **Dr. Amena Sultana**, Senior Scientific Officer, Agronomy Division, Bangladesh Rice Research Institute, for his kind cooperation, encouragement, affectionate feelings, technical help, valuable advice and helpful discussion throughout the entire period of research work and preparation of the thesis.*

*The author would like to express gratitude to **Mohammad Mosharraf Hossain**, Associate professor & Chairman, Dept. of Soil Science, Sher-e-Bangla Agricultural University, Sher-e-Bangla Nagar Dhaka. The author feels proud to express and boundless indebtedness to all the honorable teachers- Prof. Dr. Alok Kumar Paul, Prof. Dr. Assaduzzaman Khan, Prof. A.T.M Samsuddoha, Prof. Mst. Afroze Jahan, Assistant Professor ASM Fazle Bari, Department of Soil Science, Sher-e-Bangla Agricultural University, Sher-e-Bangla Nagar Dhaka, for their valuable teaching, sympathetic co-operation and inspirations throughout the course of this study.*

Cheerful acknowledgements are expressed to his elder brothers Mosharaf Hossain Munshi, to his friends Arif, Sujan, Pavel, faisal, shamol, Ramiz, Sento, Saidur, Shaon, Reza, Motahar, Shohag, Alam and relatives especially Faruk ,Yusuf, Choton, Fatema, Bithy, Moyna, Piyas, Piyal, Md.Nuruzzaman, Sumaya, Halima, Jannat, Jakia and all his well-wisher.

Finally the author is ever grateful to his respective parents Abdul Khaleque Munshi and Mrs. Monzuara Begum for their everlasting love, patience, moral and constant blessings.

The Author

EFFECT OF MODIFIED CHITOSAN ON GROWTH AND YIELD OF RICE (BRRI dhan62)

ABSTRACT

An experiment was carried out at the research field of the Department of Soil science, of Sher-e-Bangla Agricultural University, Dhaka-1207 during the period from July to December 2015 to investigate the effect of modified chitosan on growth, yield contributing characters and yield of rice cv. BRRI dhan62. Single factor experiment was laid out in a randomized complete block design (RCBD) with three replications. The experiment comprised of five levels of modified chitosan and the treatments were T_1 (0.5 t/ha modified chitosan), T_2 (1.0 t/ha modified chitosan), T_3 (2.0 t/ha modified chitosan), T_4 (4.0 t/ha modified chitosan) and T_5 (without modified chitosan or control). Different doses of modified chitosan was applied and mixed in the soil before transplanting of rice seedlings. Soil application of modified chitosan was significant on all parameters studied except plant height, panicle length, total number of grains/panicle, filled grains/panicle, unfilled grains/panicle, but the grain number increased with the increasing doses of modified chitosan among the treatments compare to the control. The experimental results revealed that 4.0 t/ha modified chitosan (T_4) produced highest grain yield (6.15 t/ha), maximum number of tillers/hill (17.85), highest straw yield (6.80 t/ha), highest biological yield (12.95 t/ha), maximum 1000-grain weight (26.90), highest value of pH (6.23), highest organic carbon content (0.97%), and highest organic matter content (1.67%). And the lowest values of these parameters were observed in the control treatment having without modified chitosan application. From the results it is concluded that almost all of growth parameters, yield and yield contributing characters and chemical properties of soil were improved due to using modified chitosan in soils compared to the control. These results suggest that modified chitosan could be used to increase the rice grain yield with sustainable soil health.

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LIST OF ACRONYMS

AEZ	Agro- Ecological Zone
Anon.	Anonymous
AIS	Agricultural Information Service
BARI	Bangladesh Agricultural Research Institute
BAU	Bangladesh Agricultural University
BBS	Bangladesh Bureau of Statistics
BINA	Bangladesh Institute of Nuclear Agriculture
BRRI	Bangladesh Rice Research Institute
cm	Centimeter
CV	Coefficient of Variance
cv.	Cultivar (s)
DAT	Days After Transplanting
^0C	Degree Centigrade
et al.	And others
FAO	Food and Agriculture Organization
g	Gram (s)
HI	Harvest Index
IRRI	International Rice Research Institute
hr	Hour(s)
K_2O	Potassium Oxide
kg	Kilogram (s)
LSD	Least Significant Difference
m	Meter
m^2	Meter squares
mm	Millimeter
MOP	Muriate of Potash
N	Nitrogen
No.	Number
NS	Non significant
%	Percentage
P_2O_5	Phosphorus Penta Oxide
S	Sulphur
SAU SRDI	Sher-e- Bangla Agricultural University Soil Resources Development Institute
t ha^{-1}	Ton per hectare
TSP	Triple Super Phosphate
var.	Variety
Wt.	Weight
Zn	Zinc

CHAPTER I

INTRODUCTION

Rice belongs to the Gramminae family with the scientific name *Oryza sativa* L. Rice is the staple food for more than three billion people that is over half of the world's total population (FAO, 2004). Rice is the most important source of the food energy for more than half of the human population. Rice is the staple food of about 135 million people of Bangladesh. It provides nearly 48% of rural employment, about two-third of total calorie supply and about one-half of the total protein intakes of an average person in the country. Rice sector contributes one-half of the agricultural GDP and one-sixth of the national income in Bangladesh. Rice is grown in 114 countries across the world on an area about 150 million hectares with annual production of over 525 million tones, constituting nearly 11 per cent of the world's cultivated land (Rai, 2006). More than 90 per cent of the world's rice is produced and consumed in Asia where it is an integral part of culture and tradition. In Asia, it is the main item of the diet of 3.5 billion people. Therefore, increase in population will require 70 percent more rice in 2025 than is consumed today (Kim and Krishnan, 2002). According to the Food and Agriculture Organization (FAO) of the U.N., 80% of the world rice production comes from 7 countries (UAE-FAO, 2012). In worldwide, 474.86 million metric tons of rice was produced from 159.64 million hectares of land with an average yield of 4.43 t/ha during the year of 2014-15 (USDA, 2015). USDA estimates Bangladesh has to produce around 34.51 million tons of rice from an estimated 11.7 million hectares of land in the year 2016-17.

Aman rice is more popular in Bangladesh. Among different groups of rice, transplant aman rice covers about 49.11% of total rice area and it contributes to 38.11% of the total rice production in the country (BBS, 2013). But rice production is affected by various biotic and abiotic constraints. Bangladesh is facing a chronic shortage of food over the year due to high population pressure. About 220 hectares agricultural lands are decreased per year due to urbanization, industrialization, housing and road construction purposes. Fifty lac acres of agricultural land decreased during last 20 years (Anon, 2007). Moreover, some rice growing area is now being used as ponds for raising fishes to meet increasing protein demand of people. Now it is essential to find out sustainable technology for poverty alleviation and ensuring food security for increasing population. So, the researchers have to think how to solve the food problem of the country. Beside, agriculture is the single largest producing sector of Bangladesh economy since it comprises about 23.08% of the country's Gross Domestic Product (GDP) (AIS, 2006) where employs around 45% of the total labor force. Despite such a steady growth in agriculture as well as in food production, Bangladesh has been facing persistent challenges in achieving food security. This is mainly due to natural disasters and fluctuations in food prices from the influence of volatile international market for basic food items (Rahman, 2011). Besides that socio-economic factor and lack of inputs also important. That is why continuous efforts are being taken towards the development of new rice cultivars and their management practices to increase the yield per unit area and meet other requirements. So, we have to think in other ways such as (i) by applying biotechnology, which is also difficult in the present condition in Bangladesh and (ii) by improving plant growth through the application of plant growth regulators like chitosan.

Chitosan is a natural biopolymer which stimulates growth and increases yield of plants as well as induces the immune system of plants (Boonlertnirun *et al.*, 2008). Plant treated with chitosan showed significantly greater number of branches/plant than untreated control (Reddy *et al*., 2000). In agriculture, chitosan is used primarily as a natural seed treatment and plant growth enhancer and as an ecologically friendly biopesticide substance that boosts the innate ability of plants to defend themselves against fungal infections (Linden *et al*., 2000). Plants with high content of chitin have better disease resistance (Khan *et al*., 2003). The natural bio-control active ingredients, chitin/chitosan are found in the shells of crustaceans, such as lobsters, crabs and shrimps and many other organisms, including insects and fungi. It is one of the most abundant biodegradable materials in the world. Degraded molecules of chitin/chitosan exist in soil and water. Chitosan has a wide scope of application on various plants. With high affinity and non-toxicity, it does no harm to human beings and livestock. Chitosan regulates the immune system of plants and induces the excretion of resistant enzymes. Moreover, chitosan not only activates the cells but also improves its disease and insect resistant ability (Doares et al., 1995). Chitosan has strong effects on agriculture such as acting as the carbon source for microbes in the soil, accelerating the transformation process of organic matter into inorganic matter and assisting the root system of plants to absorb more nutrients from the soil. Chitosan is absorbed by the roots after being decomposed by bacteria in the soil and chitin secreted by the roots (Somashekar and Ricard, 1996, Brian *et al.*, 2004). Application of chitosan in agriculture, even without chemical fertilizer can increase the microbial population by large numbers and transforms organic nutrient into inorganic nutrient which is easily absorbed by the plant roots (Bolto *et al*., 2004).

Modified chitosan, the acetylated form of chitosan powder, is the raw material prepared from the shell byproducts of crustaceans such as shrimp, crabs, lobster, cell wall of fungi etc. by the sequential process of grinding, drying and finally by sieving.

Therefore, the present study was designed to investigate the effect of various doses of modified chitosan on growth, yield and yield contributing characters of BRRI dhan62.

The objectives of the study are as follows:

i) To examine growth and yield performance of BRRI dhan62 as influenced by different levels of modified chitosan.

ii) To evaluate the doses of modified chitosan for maximizing the yield of rice.

iii) To observe the chemical properties of soil.

CHAPTER II

REVIEW OF LITERATURE

Plant growth regulators are the substances that regulate the growth of plants in an incredible form. Many scientists are now studying the pattern of growth and development of plant treated with different plant growth regulators. Chitosan is a natural biopolymer which stimulates growth and increases yield and yield contributing characteristics of various crops such as rice. Extensive studies of the regulatory effects of chitosan on various crops have been carried out worldwide by different workers and scientists. Some or the related reports are reviewed below.

2.1 Effect of chitosan application on morphological and growth characters

2.1.1. Plant height

Supachitra *et al.* (2011) conducted an experiment for determining the plant growth stimulating effects of different doses of chitosan on Thai indica rice (*Oryza sativa* L.) cv. Leung PraTew 123. Rice seedlings were treated with oligomeric chitosan with 80% degree of deacetylation at the concentration level of 40 mg/L by seed soaking overnight before sowing, followed by spraying on 2-week and 4-week old seedlings, respectively. The oligomeric chitosan stimulated plant height.

Application of oligochitosan also increased mineral nutrient uptake of coffee and stimulated the growth of coffee seedlings. Spraying oligo-chitosan with concentration of 60 mg/L stimulated and increased the height of the coffee seedlings up to 33.51% (Nguyen *et al*., 2011).

A positive and stimulating effect of chitosan was observed on the growth of roots, shoots and leaves of several crop plants (Chibu and Shibayama, 2001). Chitosan under the stress of low temperature increased shoot height and root length in maize plants compared to that of the control (Guan *et al*., 2009).

Boonlertnirun *et al.* (2008) found that application of chitosan on rice plants did not influence and stimulate the plant height significantly. Sultana (2007) applied Miyodo on rice and reported that plant height increased in Miyodo application plant than from the control.

A greenhouse experiment was conducted by Boonlertnirun *et al.* (2006) for determining the most effective chitosan dose and appropriate application method for increasing rice yield and revealed that the application of chitosan with different molecular weights and different application methods did not affect plant height.

The application of chitosan via seed soaking and spraying 4 times created variation in number of tillers/plant and dry matter accumulation, but did not affect plant height, 1000-grain weight and number of seeds/head of rice (Boonlertnirun *et al*., 2005).

Applications with different molecular weights of chitosan did not affect plant height. It was also found that plant height was not significantly different under various methods of application. Khan *et al*. (2002) which reported that foliar application of oligomeric chitosan did not affect plant height of soybean. Khan *et al.* (2002) conducted an experiment and revealed foliar application of chitosan and chitin oligomers did not affect ($p>0.05$) maize or soybean height, root length, leaf area, shoot or root or total dry mass.

Ouyang and Xu (2003) conducted a experiment with Chinese cabbage (*Brassica campestris*) cv. Dwarf hybrid No.1 and found that seed dressing with chitosan at the rate of 0.4-0.6 mg/g seed and leaf spraying 20-40 micro g/L increased plant height and leaf area of Chinese cabbage.

Vasudevan *et al.* (2002) revealed that application of chitosan formulations can increase in root and shoot length, and grain yield. It also increases in the growth of nursery-raised seedlings such as cucumber, pepper and tomato among others.

Sultana *et al.* (2015) conducted a field experiment at the Atomic Energy Research Establishment area on rice plant. Four different concentrations were used in this experiment that is 0, 40, 80 and 100 ppm oligomeric chitosan and four times foliar spray after germination were carried out. Plant height does not show any statistically significant differences between control and 40 ppm oligo-chitosan sprayed plants. But for 80 and 100 ppm oligo-chitosan sprayed rice plants show significant differences with compared to control.

2.1.2. Total number of tillers/hill

Limpanavech *et al.* (2008) conducted an experiment with six types of chitosan molecules, P-70, O-70, P-80, O-80, P-90, and O-90, for determining the effects on Dendrobium 'Eiskul' floral production. Chitosan O-80 at all concentrations tested, 1, 10, 50, and 100 mg/L could induce early flowering and increase the accumulative inflorescence number during the 68 weeks of the experimental periodic time, when compared to the non-chitosan treated controls.

The application of polymeric chitosan by seed soaking before planting followed by four foliar sprayings of chitosan doses throughout cropping season significantly increased the number of tillers per plant (Boonlertnirun *et al*., 2006).

Boonlertnirun *et al.* (2008) conducted an experiment and revealed that the maximum tiller numbers obtained from treatment of seed soaking in chitosan solution before planting and then applying in soil but however did not significantly differ from the control with no application of chitosan.

Hoque (2002) carried out an experiment on seed germination and seedling growth by seed soaking of different wheat cultivars with 0.16 ml/L, 0.33 ml/L and 0.66 ml/L of CI-IAA, GABA and TNZ-303. The number of tiller enhanced significantly at 0.33 ml/L of PGR compared to that of control.

2.2 Effect of chitosan application on yield contributing characters

2.2.1 Effective tillers/hill and length of panicle

Islam (2007) applied Miyobi on rice at the rate of 1.0, 2.0, 3.0 and 4.0 mg/L and concluded that panicle length increased with increasing hormone concentration and the highest panicle length was observed in 4.0 mg/L Miyobi application. Similar result was also reported by Sultana (2007) in rice.

Lu *et al.* (2002) studied and found that the panicle numbers of rice were increased after watering with chitosan at the rate of 0.4 g /50 cm^3 (Chitosan: water).

Hoque (2002) was conducted the field experiment and observed that the wheat cv. Treated with GABA (0.33 ml/L) produced the tallest spike (9.00 cm) followed by TNZ303 (8.10 cm) and CL-IAA (7.95 cm) respectively. The length of spike in GABA treated plant was significantly higher than from the other treatments.

Ohta *et al*. (2001) also reported that the application of a soil mixing of chitosan at the rate of 1% w/w at sowing remarkably increased flower numbers of *Eustoma grandiflorum.*

Harada *et al.* (1995) who revealed and found that soil application with chitosan increased branch and the node numbers per plant in soybean crop.

Sultana *et al*. (2015) conducted a field experiment at the Atomic Energy Research Establishment area on rice plant. Four different doses were used in this experiment that is 0, 40, 80 and 100 ppm oligomeric chitosan and four times foliar spray after germination were carried out. Finally it is observed that number of effective tillers per plant, panicle length of rice show significant differences between control plants and foliar sprayed chitosan plants.

2.2.2 Number of total, filled and unfilled grains/panicle

Boonlertnirun *et al.* (2005) indicated that seed numbers per panicle of rice plant cv. Suphan Buri-1 were not affected by various chitosan concentrations.

Sultana *et al.* (2015) conducted a field experiment on rice plant. Four different concentrations were used in this experiment that is 0, 40, 80 and 100 ppm oligomeric chitosan and four times foliar spray after germination were carried out. Finally it is observed total grains per panicle of rice show significant differences between control plants and foliar sprayed chitosan plants.

Boonlertnirun *et al.* (2006) conducted a pot experiment in a greenhouse and the results indicated that application of polymeric chitosan by seed soaking before planting followed by four foliar sprayings of chitosan doses throughout cropping season did not affect filled grains/panicle. Application of polymeric chitosan significantly resulted in less number of unfilled grains than those of the others. The application of polymeric chitosan by seed soaking and four foliar sprayings throughout cropping season reduced unfilled grains of rice.

Utsunomiya *et al*. (1998) reported that the number of flowers and the harvesting fruits of purple passion fruit increased with soil treated with oligomeric chitosan under high nitrogen conditions.

Boonlertnirun *et al.* (2008) conducted an experiment with 4 treatments. The treatments were T_1- no chitosan application (control), T_2- seed soaking with chitosan solution, T_3 - seed soaking and soil application with chitosan solution and T_4 - seed soaking and foliar spraying with chitosan solution. Application of chitosan by seed soaking in chitosan solution before planting and then applying in soil tended to produce more filled grains/panicle than the other methods but it was not significantly different from the other treatments and the control.

Kananont *et al*. (2015) conducted an experiment with Fermented chitin waste (FCW) with three levels of FCW @ (0.25%, 0.50% and 1.0% (w/w)) along with CF = soil supplemented with chemical fertilizer and CMF = soil supplemented with chicken manure fertilizer. The results found that FCW @ 1% the filled grains/panicle differ significantly from 0.5% FCW, 0.25% FCW and the rest of the treatment.

2.2.3 Thousand grain weight

An experimental trial was carried out in the two successive seasons of 2008 and 2009 to investigate and compare the enhancing effects of three different bio stimulation compounds on growth and production of sweet pepper plants. Three weeks after transplanting, plants were sprayed with any of the individual chitosan (2, 4 and 6 cm/L). Data showed that individual fruit weight and number of fruits were also improved (Ghoname *et al*., 2010).

Islam (2007) conducted a field experiment on rice. He sprayed Myobi on the rice plants @2, 3 and 4 mg/L and GABA at 2, 3 and 4 mg/L as foliar application. He observed highest 1000-seed weight for 2 mg/L GABA followed by 2 mg/L Myobi.

A greenhouse experiments were conducted to determine the effect of chitosan on drought recovery and grain yield of rice under drought conditions. Results revealed that the chitosan application before drought treatment gave the highest 1000-grain weight and also showed good recovery on yield (Boonlertnirun *et al*., 2007).

Boonlertnirun *et al.* (2006) conducted a pot experiment in a greenhouse with application method of chitosan as seed soaking; seed soaking + foliar spraying and foliar spray; and the type of chitosan as polymeric chitosan, oligomeric chitosan and monomeric chitosan. The results revealed that the application of chitosan with different molecular weights and different application methods did not affect thousand grain weights.

Krivtsov *et al*. (1996) found that thousand grain weight and spike weight of wheat plants increased in treatment with low concentrations of polymeric chitosan than the other treatment.

Boonlertnirun *et al.* (2008) conducted an experiment with 4 treatments. The treatments were T_1- no chitosan application (control), T_2- seed soaking with chitosan solution, T_3 - seed soaking and soil application with chitosan solution and T_4 - seed soaking and foliar spraying with chitosan solution. The results revealed that application of chitosan by varying application methods did not affect 1,000- grain weight (gm).

Kananont *et al*. (2015) conducted an experiment with Fermented chitin waste (FCW) with three levels of FCW @ (0.25%, 0.50% and 1.0% (w/w)) along with CF = soil supplemented with chemical fertilizer and CMF = soil supplemented with chicken manure fertilizer. The results found that FCW @ 0.50% increases 1000 grain weight (gm) significantly from the rest of the treatment.

Sultana *et al.* (2015) conducted a field experiment on rice plant. Four different concentrations were used in this experiment that is 0, 40, 80 and 100 ppm oligomeric chitosan and four times foliar spray after germination were carried out. Finally it is observed that 1000 grain weight (gm) show significant differences between control plants and foliar sprayed chitosan plants.

2.2.4 Straw yield

Sultana *et al.* (2015) conducted a field experiment on rice plant. Four different concentrations were used in this experiment that is 0, 40, 80 and 100 ppm oligomeric chitosan and four times foliar spray after germination were carried out. Finally it is observed that straw yield show significant differences between control plants and foliar sprayed chitosan plants and highest straw yield was observed under 100 ppm oligomeric chitosan and lowest straw yield was observed under 0 ppm oligomeric chitosan.

Kananont *et al*. (2015) conducted an experiment with Fermented chitin waste (FCW) with three levels of FCW @ (0.25%, 0.50% and 1.0% (w/w)) along with CF = soil supplemented with chemical fertilizer and CMF = soil supplemented with chicken manure fertilizer. The results found that FCW @ 1% the straw yield differ significantly from 0.5% FCW, 0.25% FCW and the rest of the treatment.

2.2.4 Grain yield

Sultana (2010) from BAEC, Bangladesh reported that the oligochitosan was applied for its potential use as plant growth promoter on growth and productivity of Maize (*Zea mays.* L) Plants. The foliar spraying of oligochitosan (molecular weight 7,000 Da) with the concentration of 25, 50 and 75 mg/L was applied. The results showed that the application of oligochitosan, at the concentration of 75 mg/L, plays a significant role in terms of weight of cob and weight of seeds per Maize and ultimately maize yield.

Abdel-Mawgoud *et al*., (2010) found an improvement effects on strawberry production or yield as a result of chitosan application on strawberry plant.

Krivtsov *et al.* (1996) revealed that wheat plants treated with polymeric or oligomeric chitosan increased grain yield. In contrast, the work of Kuznia *et al.* (1993) found that seeds of white lupine treated with chitosan doses did not consistently increase yields.

Boonlertnirun, *et al.* (2006) reported that rice yield of cultivar SuphanBuri 1 was significantly increased over the control (no chitosan) after application of polymeric chitosan at the concentration of 20 ppm.

Boonlertnirun *et al*. (2008) conducted an experiment on application of chitosan in rice production with 4 treatments. The treatments were T_1- no chitosan application (control), T_2- seed soaking with chitosan solution, T_3- seed soaking and soil application with chitosan solution and T_4- seed soaking and foliar spraying with chitosan solution. The results showed that application of chitosan by seed soaking and then soil application of chitosan significantly increased rice yield over the other treatments.

A Greenhouse experiments were conducted for determining the effect of chitosan on drought recovery and grain yield of rice under drought conditions. Results revealed that the chitosan applied before drought treatment gave the highest yield and yield components and also showed good recovery, Boonlertnirun *et al*. (2007).

Sultana *et al.* (2015) conducted a field experiment on rice plant. Four different concentrations were used in this experiment that is 0, 40, 80 and 100 ppm oligomeric chitosan and four times foliar spray after germination were carried out. Finally it is observed that grain yield show significant differences between control plants and foliar sprayed chitosan plants.

Kananont *et al*. (2015) conducted an experiment with Fermented chitin waste (FCW) with three levels of FCW @ (0.25%, 0.50% and 1.0% (w/w)) along with CF = soil supplemented with chemical fertilizer and CMF = soil supplemented with chicken manure fertilizer. The results found that FCW @ 1% the grain yield differ significantly from 0.5% FCW, 0.25% FCW and the rest of the treatment.

Chitin or derivatives chitosan level improves the durability and resistance of the plant, makes it not easily infected by germs, not proliferate even infected, and cures the disease by itself. Using chitosan in agriculture with less use of chemical fertilizer increases the production, in different kinds of plant, by 15-20% (Hong *et al*., 1998).

2.3 Effect of Chitosan application on chemical properties (pH, organic carbon and organic matter) of soil

Fermented chitin waste (FCW), a by-product obtained from chitinase production using chitin fermentation. Kananont *et al*. (2015) conducted an experiment with Fermented chitin waste (FCW) with three levels of FCW @ (0.25%, 0.50% and 1.0%

(w/w)) along with CF = soil supplemented with chemical fertilizer and CMF = soil supplemented with chicken manure fertilizer. The results found that FCW @ 1% the pH differ significantly from 0.5% FCW, 0.25% FCW and the rest of the treatment. The pH ranges from 5.01 to 5.93 among the treatment. The highest pH was obtained (5.93) with FCW @ 1% and lowest pH was obtained (5.01) with CF = soil supplemented with chemical fertilizer.

Application of FCW to the soil led to an increased organic carbon level in the soil. The organic carbon level differs significantly among the treatments. The results found that FCW @ 1% the organic carbon differ significantly from 0.5% FCW, 0.25% FCW and the rest of the treatment.

Application of FCW to the soil also led to an increased organic matter in the soil. OM improves the physical, chemical and biological properties of soil, as well as giving a better soil aggregation, available water content and enhanced cation exchange capacity, leading to improved soil fertility. (Khaleel *et al*., 1981, Metzger and Yaron, 1987, Matsumoto *et al.,* 1999, Farooq and Nawaz, 2014). The organic matter differs significantly among the treatments. The results found that FCW @ 1% the organic matter differ significantly from 0.5% FCW, 0.25% FCW and the rest of the treatment. Organic matter level ranges from 1.82 to 2.35 among the treatment. The highest organic matter level was obtained (2.35) with FCW @ 1% and lowest organic matter level was obtained (1.82) with CF = soil supplemented with chemical fertilizer.

CHAPTER III

MATERIALS AND METHODS

The experiment was conducted at the soil science field of Sher-e-Bangla Agricultural University, Dhaka-1207 during the period from July to December 2015. This chapter deals with a brief description on experimental site, climate, soil, and land preparation, layout of the experimental design, intercultural operations, data recording and their analyses.

3.1 Site description

3.1.1 Geographical location

The experimental area was situated at 23°77′N latitude and 90°33′E longitude at an altitude of 8.6 meter above sea level.

3.1.2 Agro-ecological region

The experimental field belongs to the Agro-ecological zone of "The Modhupur Tract", AEZ-28 (Anon., 1988a). This was a region of complex relief and soils developed over the Modhupur clay, where floodplain sediments buried the dissected edges of the Modhupur Tract leaving small hillocks of red soils as 'islands' surrounded by floodplain (Anon., 1988b). The experimental site was shown in the map of AEZ of Bangladesh in Appendix I.

3.1.3 Climate

The experimental area is under the sub-tropical climate that is characterized by high temperature, high humidity and heavy rainfall with occasional gusty winds in kharif season (April-September) and less rainfall associated with moderately low temperature during the Rabi season (October-March). The weather data during the study period of the experimental site is shown in Appendix II.

3.1.4 Soil

The farm belongs to the general soil type, Shallow Red Brown Terrace soils under Tejgaon Series. Top soils were clay loam in texture, olive-gray with common fine to medium distinct dark yellowish brown mottles. Soil pH was 5.43 and had organic carbon 0.60% and organic matter content is 1.03%. The experimental area was flat having available irrigation and drainage system. The land was above flood level and sufficient sunshine was available during the experimental period. Soil samples from 0-15 cm depths were collected from experimental field. The chemical analyses were done in the laboratory of the Department of Soil Science of Sher-e-Bangla Agricultural University, Dhaka-1207. The physicochemical properties of the soil are presented below.

Table 1: Morphological characteristics of the soil of experimental field

Morphological features	Characteristics
Location	Soil science field ,SAU, Dhaka
AEZ	Madhupur field (28)
General soil type	Shallow red brown terrace soil
Land type	High land
Soil series	Tejgaon
Topography	Fairly leveled
Flood level	Above flood level
Drainage	Well drained

Table 2: Physical and chemical properties of the initial soil

Characteristics	Value
% Sand	27
% Silt	43
% Clay	30
Textural class	Silty-clay
pH	5.43
Organic carbon (%)	0.60
Organic matter (%)	1.03
Total N (%)	0.03
Available P (ppm)	21.00
Exchangeable K (me/100 g soil)	0.12
Available S (ppm)	48

3.2 Experiment details

3.2.1 Crop/Planting material

BRRI dhan62 were used as the test crop. The variety BRRI dhan62 was developed by Bangladesh Rice Research Institute (BRRI), Joydebpur, Gajipur, Bangladesh for T. Aman season. The pedigree line of BRRI dhan62 is BR 517-2R-27-3. Average height of the plant is 98 cm. The grains are long, slender and white in color. The grains are zinc enriched. The growth duration is about 100 day.

3.2.2 Experimental Treatments

The single factor experiment was compared with five treatments of chitosan.

T_1= 0.5 t/ha modified chitosan

T_2= 1.0 t/ha modified chitosan

T_3= 2.0 t/ha modified chitosan

T_4= 4.0 t/ha modified chitosan

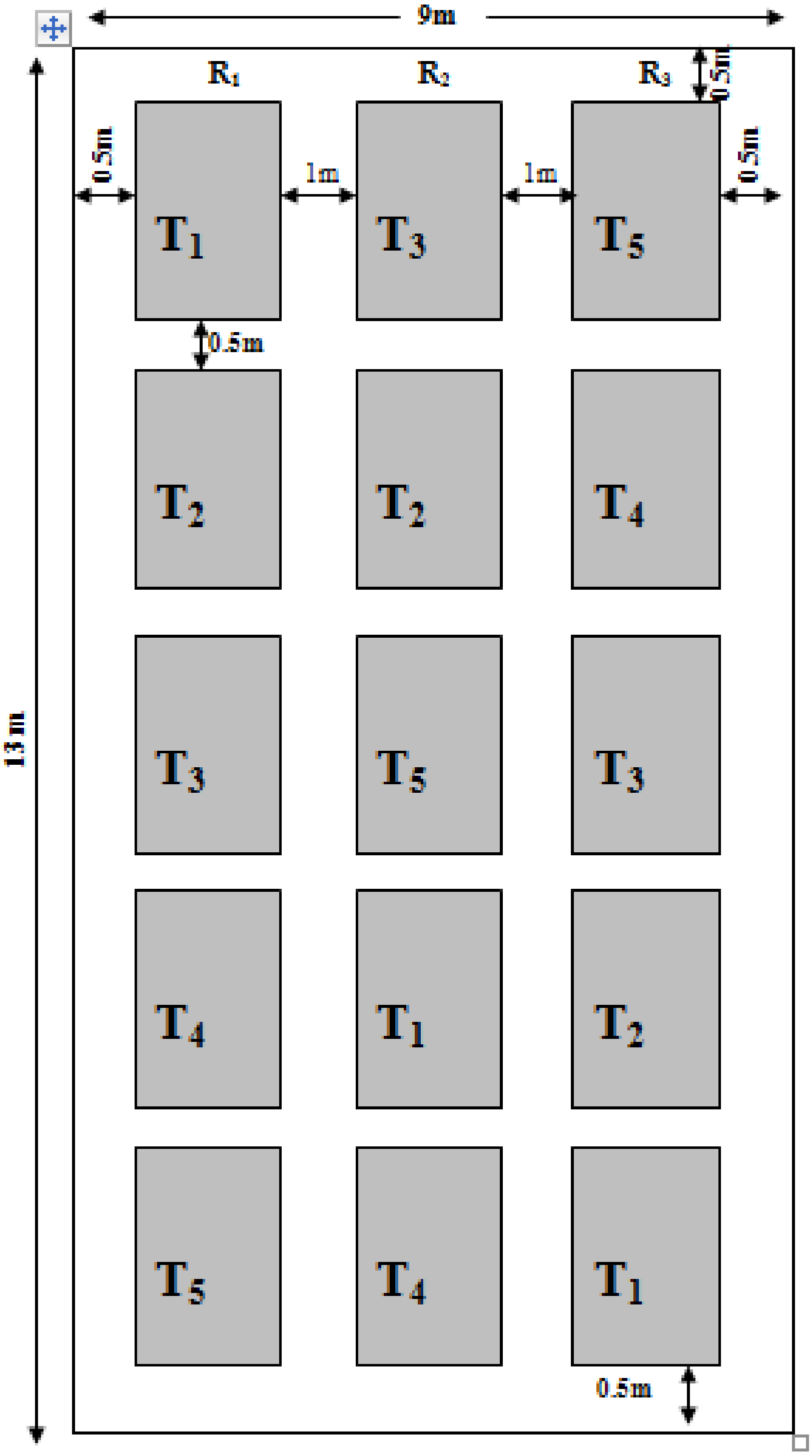
T_5= without modified chitosan (Control)

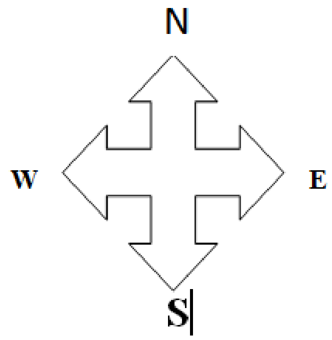
3.2.3 Preparation of modified chitosan

Modified chitosan was prepared using shrimp shell byproducts collecting from the Khulna region of Bangladesh following a new traditional method. The prepared modified chitosan was used in the experiment during the final land preparation.

3.2.4 Experimental design

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 2 m × 2 m. Total plots in the experimental field were 15. 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. The layout of the experiment has been shown in Fig 1.





Distance between replications: 1m

Distance between treatments: 0.5m

Plot size: 2m×2m = 4m^2

Total length of the field:
(5×2m) + (6×0.5m) = 13m

Total width of the field:
(3×2m)+(0.5m+1m+1m+0.5m= 9m

Total area of the field:
L×B = 13m × 9m = 117m^2

<u>Replications:</u>

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

<u>Treatment Combinations:</u>

T_1= 0.5 t/ha modified Chitosan
T_2= 1.0 t/ha modified Chitosan
T_3= 2.0 t/ha modified Chitosan
T_4= 4.0 t/ha modified Chitosan
T_5= 0.0 t/ha modified Chitosan

Fig: The layout of the experimental field

3.3 Growing of crops

3.3.1 Seed collection and sprouting

Seeds BRRI dhan62 were collected from BRRI, Joydebpur, Gazipur, Bangladesh. Healthy seeds were selected following standard method. Seeds were immersed in water in a bucket for 24 hrs. These were then taken out of water and tightly kept in gunny bags. The seeds started to sprout after 48 hrs which became ready for sowing in 72 hrs.

3.3.2 Raising of seedlings

A common procedure was followed in rising of seedlings in the nursery bed. The nursery bed was prepared by puddling with repeated ploughing followed by laddering. Weeds were removed and irrigation was gently provided to the bed as and when needed. No fertilizer was used in the nursery bed.

3.3.3 Seed sowing

Seeds were sown on the nursery bed on August 3, 2015 for raising nursery seedlings.

3.3.4 Preparation of experimental land

The experimental field was first opened on August 20, 2015 with the help of a tractor drawn disc plough; later on August 22, 2015 the land was irrigated and prepared by three successive ploughing and cross ploughing with a tractor plough and subsequently leveled by laddering. All kinds of weeds and residues of previous crop were removed from the field. After the final land preparation the field layout was made on August 24, 2015 according to experimental plan. Individual plots were cleaned and finally leveled with the help of wooden plank so that no water pocket could remain in the puddle field.

3.3.5 Fertilizer dose and methods of application

Unit plots of the experiment were fertilized with 150, 58, 58, 38 and 10 kg/ha of urea, triple super phosphate (TSP), muriate of potash (MOP), gypsum and zinc sulphate respectively. The entire amounts of triple super phosphate, muriate of potash, gypsum and zinc sulphate were applied as basal dose at the time of transplanting of seedlings. Urea was top-dressed in three equal splits. The first one-third urea was top-dressed after seedling recovery, second during the vegetation stage and third at 7 days before panicle initiation.

3.3.6 Transplanting of seedlings

25 days old seedlings were uprooted carefully from the nursery beds on 26 August, 2015 for transplantation. For this purpose the nursery beds were made wet by the application of water in previous day before uprooting the seedlings to minimize mechanical injury of roots.

3.3.7 Intercultural operations

3.3.7.1 Gap filling

After one week of transplantation, a minor gap filling was done as and where necessary using the seedling or separated tillers from the previous source as per treatment.

3.3.7.2 Weeding

The crop was infested with some weeds during the early stage of crop establishment. Two hand weeding were done for each treatment; first weeding was done at 20 days after transplanting followed by second weeding at 15 days after first weeding.

3.3.7.3 Application of irrigation water

Irrigation water was added to each plot according to the need. All the plots were kept irrigated maintaining 3-5 cm stagnant water throughout the entire period up to 15 days before harvesting.

3.3.7.4 Plant protection measures

Plants were infested with rice stem borer (*Scirphophaga incertolus*) and leaf hopper (*Nephotettix nigropictus*) to some extent which were successfully controlled by applying Diazinon @ 10 ml/10 liter of water for 5 decimal lands and by Ripcord @ 10 ml/10 liter of water for 5 decimal lands as and when needed. Crop was protected from birds during the grain filling period. For controlling the birds watch man was deep laid, especially during morning and afternoon.

3. 4 General observation of the experimental field

The field was investigated time to time to detect visual difference among the treatment and any kind of infestation by weeds, insects and diseases so that considerable losses by pest could be minimized. The field looked nice with normal green color plants. Incidence of stem borer, green leaf hopper, leaf roller and rice hispa was observed during tillering stage that controlled properly. No bacterial and fungal disease was observed in the field.

3.5 Harvesting and post harvest operation

Maturity of crop was determined when 90% of the grains became golden yellow in color. Harvesting was done on November 20, 2015. Five hills per plot were preselected randomly from which different growth and yield attributes data were collected and 1m^2 areas from middle portion of each plot was separately harvested and bundled, properly tagged and then brought to the threshing floor for recording grain and straw yield. Threshing was done by using pedal thresher. The grains were cleaned and sun dried to a moisture content of 14 % approximately. Straw was also sun dried properly.

3.6 Recording of data

The followings data were recorded during the experiment.

A. Crop growth characters

i. Plant height (cm) at harvest

ii. Number of tillers/hill

B. Yield and yield components

i. Number of effective tillers/hill

ii. Number of non effective tillers/hill

iii. Length of panicle (cm)

iv. Number of filled grains/panicle

v. Number of unfilled grains/panicle

vi. Number of total grains/panicle

vii. Weight of 1000 grains (g)

viii. Grain yield (t/ha)

ix. Straw yield (t/ha)

x. Harvest index (%)

3.7 Detailed procedures of recording data

A. Crop growth characters

i. Plant height (cm) at harvest

Plant height was measured from the ground level to the top of the tallest panicle. Plants of 5 hills were measured and averaged for each plot

ii. Number of tillers/hill

Number of tillers/hill were counted at harvest from ten randomly pre-selected hills and averaged as their number/hill. Only those tillers having three or more leaves were considered for counting.

B. Yield and other crop characters

i. Effective tillers/hill (no.)

The panicles which had at least one grain was considered as effective tillers. The number of effective tillers/hill was recorded and finally averaged for counting effective tillers number /hill.

ii. Non effective tillers/hill (no.)

The tiller having no panicle was regarded as ineffective tillers. The number of ineffective tillers/hill was recorded and finally averaged for counting ineffective tillers number/m^2.

iii. Panicle length (cm)

Measurement of panicle length was taken from basal node of the rachis to apex of each panicle. Each observation was an average of 5 panicles.

iv. Filled grains/panicle (no.)

Grain was considered to be filled if any kernel was present there in. The number of total filled grains present on five panicles were recorded and finally averaged.

vi. Unfilled grains/panicle (no.)

Unfilled grains means the absence of any kernel inside and such grains present on each of five panicles were counted and finally averaged.

vii. Total grains/panicle (no.)

The number of filled grains/panicle plus the number of unfilled grains/panicle gave the total number of grains/panicle.

viii. Weight of 1000 grains (g)

One thousand cleaned dried grains were counted randomly from each sample and weighed by using a digital electric balance at the stage the grain retained about 12% moisture and the mean weight were expressed in gram.

ix. Grain yield (t/ha)

Grain yield was determined from the central $1m^2$ areas of each plot and expressed as t/ha on about 12% moisture basis. Grain moisture content was measured by using a digital moisture tester.

x. Straw yield (t/ha)

Straw yield was determined from the central 1m^2 areas of each plot. After separating of grains, the sub-samples were oven dried to a constant weight and finally converted to t/ha.

xi. Biological yield (t/ha)

Grain yield and straw yield were all together regarded as biological yield. Biological yield was calculated with the following formula:

Biological yield (t/ha) = Grain yield (t/ha) + Straw yield (t/ha)

xii. Harvest Index (%)

It denotes the ratio of economic yield to biological yield and was calculated with following formula.

$$\text{Harvest index (\%)} = \frac{\text{Grain yield}}{\text{Biological yield}} \times 100$$

3.8 Chemical analysis of soil samples

Soil samples were analyzed for both physical and chemical properties in the laboratory of Department of Soil Science of Sher-e-Bangla Agricultural University, Dhaka-1207.The properties studied included texture, pH, organic matter etc. The physical and chemical properties of initial soil have been presented in Table 1 and 2. The soil was analyzed following standard methods:

Particle-size analysis of soil was done by Hydrometer method (Bouyoucos, 1926) and the textural class was determined by plotting the values for % sand, % silt and % clay to the "Marshall's Textural triangular coordinate" following the USDA system.

Soil pH was measured with the help of a glass electrode pH meter using soil suspension of 1:2.5 as described by Jackson (1962).

Organic carbon in soil was determined by wet oxidation method of Walkley and Black (1934). The underlying principle is to oxidize the organic carbon with an excess of 1N $K_2Cr_2O_7$ in presence of conc. H_2SO_4 and to titrate the residual $K_2Cr_2O_7$ solution with 1N $FeSO_4$ solution. To obtain the organic matter content, the amount of organic carbon was multiplied by the Van Bemmelen factor, 1.73. The result was expressed in percentage.

3.9 Statistical Analysis

The collected data were analyzed statistically following the analysis of variance (ANOVA) technique and the mean differences were adjudged by LSD test using the statistical computer package program, Statistix 10.

CHAPTER IV

RESULT AND DISCUSSION

This chapter comprises of the presentation and discussion of the results obtained due to application of different rate of modified chitosan on growth and yield of T. Aman rice (cv. BRRI Dhan62) and chemical properties of the soils. The results of the present investigation have been presented, discussed and compared as far as available with the results of the researchers.

4.1 Plant height (cm)

Plant height was found to be statistically insignificant in all of the treatments used in the experiment. The maximum plant height (95.60 cm) was obtained in the T_3 treatment having 2.0 t/ha modified chitosan and minimum plant height (93.18) was obtained in the T_4 treatment having 4.0 t/ha modified chitosan (Table 1). According to the plant height the treatments may be arranged as $T_3>T_5>T_2>T_1>T_4$. These results were supported by Boonlertnirun *et al.* (2008) who found that application of chitosan did not influence and/or stimulate the rice plant height significantly.

Table 1. Effects of different doses of modified chitosan on plant height (cm) of T. Aman rice (BRRI dhan62) at harvest. SAU, 2015.

Treatment (dose)	Plant height (cm)
T_1	93.57
T_2	94.52
T_3	95.60
T_4	93.19
T_5	94.87
CV (%)	3.65
Level of significance	NS

T_1= 0.5 t/ha modified Chitosan T_2= 1.0 t/ha modified Chitosan

T_3= 2.0 t/ha modified Chitosan T_4= 4.0 t/ha modified Chitosan

T_5= without modified Chitosan

4.2 Total tillers/hill

The effect of different treatments on total tillers/hill was statistically significant (Figure 1). The maximum number of total tillers/hill (18.83) was obtained in the T_4 (4.0 t/ha modified chitosan) treatment which was significantly greater than that obtained in the T_5 control (without modified chitosan) and T_1 treatment (0.5 t/ha modified chitosan) and statistically identical with the treatments T_2 (1.0 t/ha modified chitosan) and T_3 (2.0 t/ha modified chitosan). However the total tillers/hill did not differ significantly in T_2, T_3, T_4 treatments. The minimum number of total tillers/hill (15.28) was obtained in the T_5 (without modified chitosan) control treatment. In producing total number of tillers/hill the treatments may be arranged as $T_4>T_3>T_2>T_1>T_5$. It was observed that total tillers/hill increased with increasing the of modified chitosan application in soil. These results were supported by Boonlertnirun *et al.* (2006) who conducted an experiment with different application method of chitosan and different molecular weights of chitosan. They found that tiller numbers per plant significantly increased with the increasing molecular weights of chitosan.

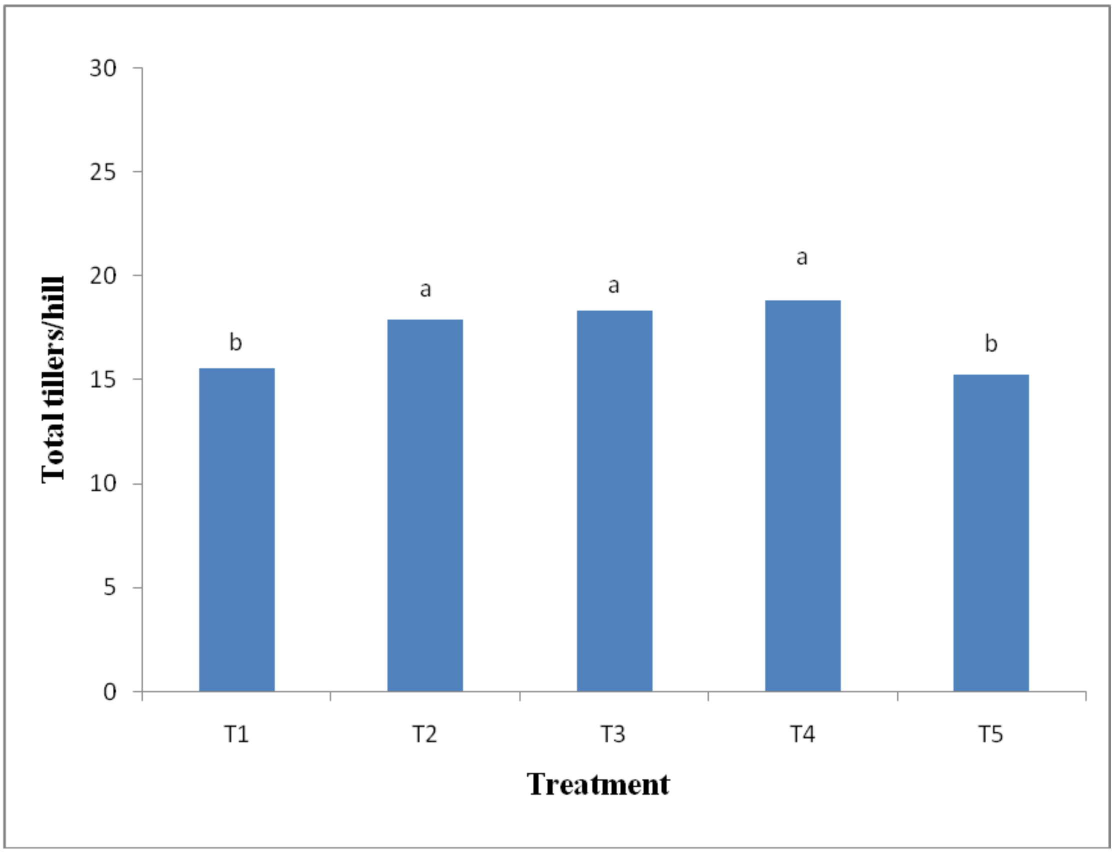


Figure 1. Effects of different doses of modified chitosan on total tillers/hill of T. Aman rice (BRRI dhan62). Mean was calculated from three replicates for each treatment. Bars with different letters are significantly different at $p \leq 0.05$ applying DMRT. SAU, 2015.

T_1= 0.5 t/ha modified chitosan

T_2= 1.0 t/ha modified chitosan

T_3= 2.0 t/ha modified chitosan

T_4= 4.0 t/ha modified chitosan

T_5= without modified chitosan

4.3 Effective tillers/hill

Figure 2 shows the effects of different treatments on effective tillers/hill. It was found that effective tillers/hill statistically significant. The highest number of effective tillers/hill (17.853) was obtained in the T_4 (4.0 t/ha modified chitosan) treatment which was significantly greater than that obtained in the T_5 control (without modified chitosan) and T_1 treatment (0.5 t/ha modified chitosan) and statistically identical to T_2 (1.0 t/ha modified chitosan) and T_3 (2.0 t/ha modified chitosan) treatment. However the effective tillers/hill did not differ significantly in T_2, T_3, T_4 treatments. The lowest number of effective tillers/hill (15.087) was obtained in the T_5 (without modified chitosan) control treatment. In producing effective number of tillers/hill the treatments may be arranged as $T_4>T_3>T_2>T_1>T_5$. It was observed that, as the rate of modified chitosan application in soil increases the effective tillers/hill also increases. These results were supported by Sultana *et al*. (2015) who conducted a field experiment with Four different concentrations that is 0, 40, 80 and 100 ppm oligomeric chitosan and four times foliar spray after germination were carried out. Finally it is observed that number of effective tillers per plant of rice show significant differences between control plants and foliar sprayed chitosan plants.

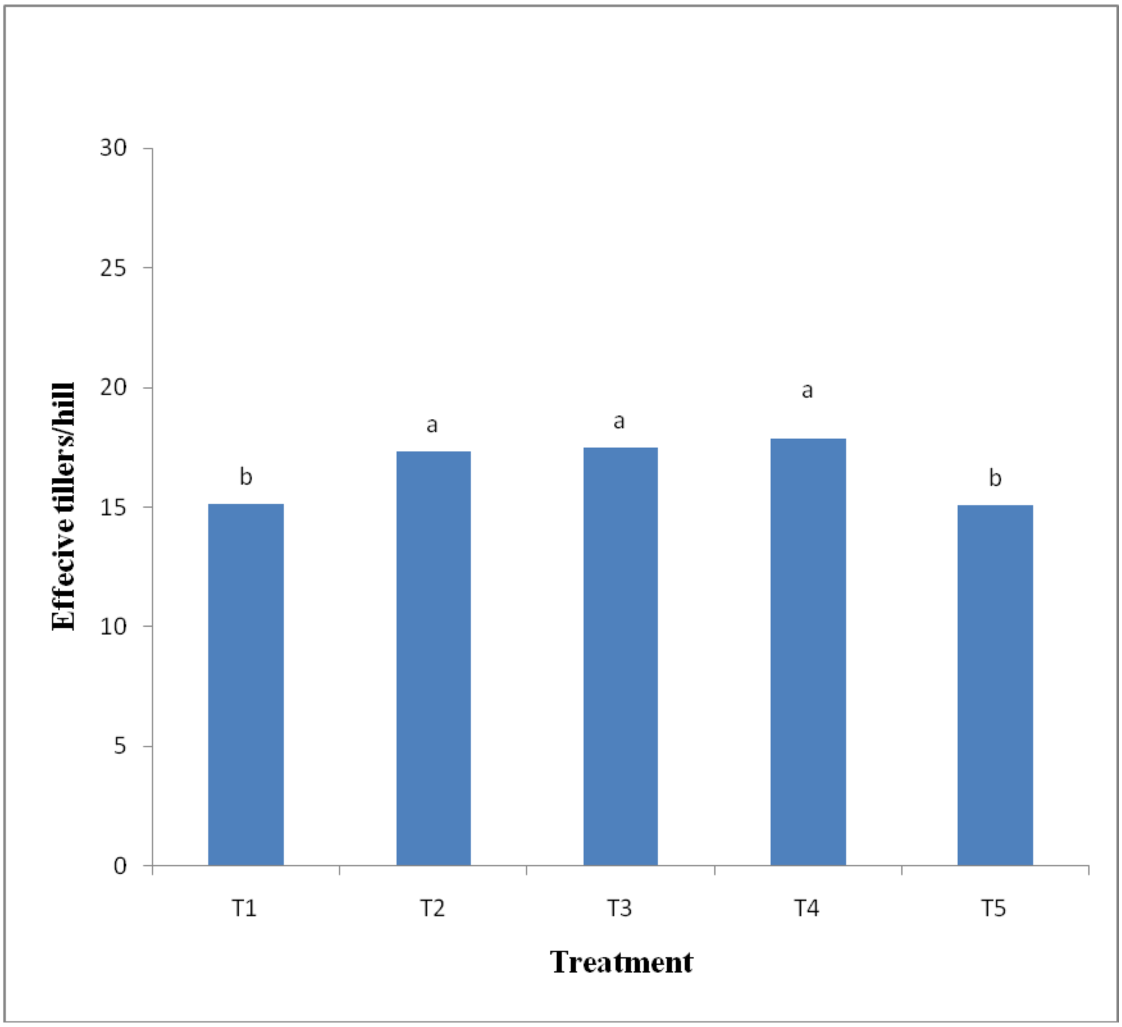


Figure 2. Effects of different doses of modified chitosan on effective tillers/hill of T. Aman rice (BRRI dhan62). Mean was calculated from three replicates for each treatment. Bars with different letters are significantly different at $p \leq 0.05$ applying DMRT. SAU, 2015.

T_1= 0.5 t/ha modified Chitosan T_2= 1.0 t/ha modified Chitosan

T_3= 2.0 t/ha modified Chitosan T_4= 4.0 t/ha modified Chitosan

T_5= without modified Chitosan

4.4 Non effective tillers/hill

The non-effective tillers/hill varied significantly due to the effects of different treatments (Fig 3). The highest non effective tillers/hill was produced (0.97) by the T_4 treatment (4.0 t/ha modified chitosan) and the lowest non effective tillers/hill was produced (0.19) by the T_5 (without modified chitosan) control treatment. In producing non effective number of tillers/hill the treatments may be arranged as $T_4>T_3>T_2>T_1>T_5$. From this study it was observed that, as the rate of modified chitosan application in soil increases the non effective tillers/hill also increases.

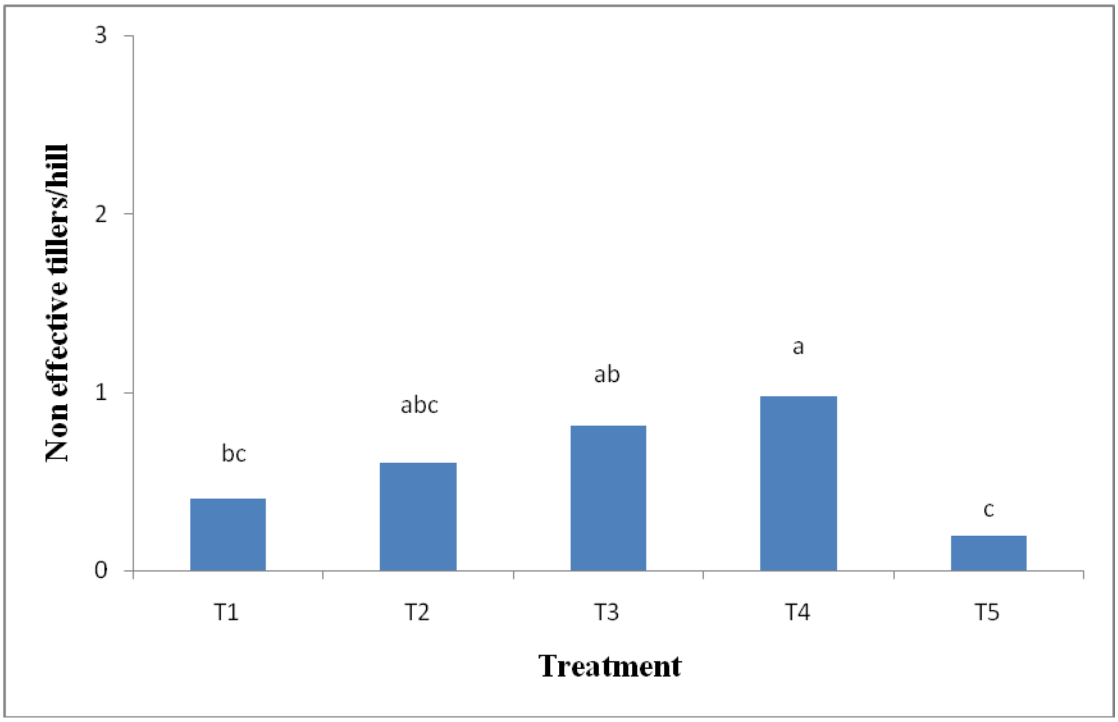


Figure 3. Effects of different doses of modified chitosan on Non effective tillers/hill of T. Aman rice (BRRI dhan62). Mean was calculated from three replicates for each treatment. Bars with different letters are significantly different at *p* ≤ 0.05 applying DMRT. SAU, 2015.

T_1= 0.5 t/ha modified Chitosan T_2= 1.0 t/ha modified Chitosan

T_3= 2.0 t/ha modified Chitosan T_4= 4.0 t/ha modified Chitosan

T_5= without modified Chitosan

4.5 Panicle length (cm)

Panicle length was found to be statistically insignificant in all of the treatments used in the experiment (Table 2). Panicle length was not influenced significantly although there was some apparent difference in panicle length in different treatments. The maximum panicle length (24.49 cm) was obtained in the T_3 treatment (2.0 t/ha modified chitosan) and minimum panicle length (23.48) was obtained in the T_1 treatment (0.5 t/ha modified chitosan) (Table 2). According to the panicle length the treatments may be arranged as $T_3>T_4>T_2>T_5>T_1$.

Table 2. Effects of different doses of modified chitosan on panicle length (cm) of T. Aman rice (BRRI dhan62) at harvest. SAU, 2015.

Treatment (dose)	Panicle length (cm)
T_1	23.48
T_2	23.73
T_3	24.49
T_4	24.15
T_5	23.50
CV (%)	3.44
Level of significance	NS

T_1= 0.5 t/ha modified Chitosan T_2= 1.0 t/ha modified Chitosan

T_3= 2.0 t/ha modified Chitosan T_4= 4.0 t/ha modified Chitosan

T_5= without modified Chitosan

4.6 Number of total grains/panicle

The results (Table 3) indicated that the effects of different treatments on total grains/panicle were found to be statistically insignificant in all of the treatments used in the experiment. Total grains/panicle was not influenced significantly although there was some apparent difference in total grains/panicle in different treatments. The maximum total grains/panicle (108.90) was obtained in the T_4 treatment (4.0 t/ha modified chitosan) and minimum total grains/panicle (98.77) was obtained in the T_5 control treatment (without modified chitosan). According to the total grains/panicle the treatments may be arranged as $T_4>T_3>T_2>T_1>T_5$. These results were supported by Boonlertnirun *et al.* (2005) indicated that seed numbers per panicle of rice plant cv. Suphan Buri-1 were not affected by various chitosan concentrations.

4.7 Number of filled grains/panicle

Table 3 reveals that the effects of different treatments on filled grains/panicle were found to be statistically insignificant in all of the treatments. Filled grains/panicle was not influenced significantly although there was some apparent difference in filled grains/panicle in different treatments. The highest number of filled grains/panicle (98.70) was obtained in the T_4 treatment (4.0 t/ha modified chitosan) and lowest number of filled grains/panicle (87.90) was obtained in the T_5 control treatment (without modified chitosan). According to the filled grains/panicle the treatments may be arranged as $T_4>T_3>T_2>T_1>T_5$.These results were supported by Boonlertnirun *et al.* (2008) who conducted an experiment with 4 treatments of chitosan and found that application of chitosan by seed soaking in chitosan solution before planting and then applying in soil tended to produce more filled grains/panicle than the other methods but it was not significantly different from the other treatments and the control.

4.8 Number of unfilled grains/panicle

Table 3 results showed that the effects of different treatments on unfilled grains/panicle were found to be statistically insignificant in all of the treatments. Unfilled grains/panicle was not influenced significantly although there was some apparent difference in unfilled grains/panicle in different treatments. The highest number of unfilled grains/panicle (13.13) was obtained in the T_3 treatment (2.0 t/ha modified chitosan) and lowest number of unfilled grains/panicle (10.20) was obtained in the T_5 control treatment (without modified chitosan). According to the unfilled grains/panicle the treatments may be arranged as $T_3>T_1>T_2>T_5>T_4$.

4.9 1000-grain weight

Figure 4 shows the effects of different treatments on 1000-grain weight. It was found that 1000-grain weight statistically significant. The highest 1000-grain weight (26.90) was obtained in the T_4 (4.0 t/ha modified chitosan) treatment which was significantly greater than that obtained in the T_5 control (without modified chitosan) treatment and statistically identical toT_1 (0.5 t/ha modified chitosan), T_2 (1.0 t/ha modified chitosan) and T_3 (2.0 t/ha modified chitosan) treatment. However the 1000-grain weight did not differ significantly in T_1, T_2, T_3, T_4 treatments. The lowest 1000-grain weight (23.26) was obtained in the T_5 (without modified chitosan) control treatment. In case of 1000-grain weight the treatments may be arranged as $T_4>T_3>T_1>T_2>T_5$. It was observed that, as the rate of modified chitosan application in soil increases the 1000-grain weight also increases. These results were supported by Boonlertnirun *et al.* (2007) by which a greenhouse experiments were conducted to determine the effect of chitosan on rice under drought conditions. Results revealed that the chitosan application before drought treatment gave the highest 1000-grain weight. Similar results were also found by Krivtsov *et al*. (1996).

Table 3. Effects of different doses of modified chitosan on yield contributing characters of T. Aman rice (BRRI dhan62) at harvest. SAU, 2015.

Treatment (dose)	Total grains panicle^{-1}	Filled grains panicle^{-1}	Unfilled grains panicle^{-1}
T_1	104.07	91.47	12.60
T_2	104.60	93.20	11.40
T_3	108.87	95.73	13.13
T_4	108.90	98.70	10.20
T_5	98.77	87.90	10.87
CV (%)	9.81	9.05	35.89
Level of significance	NS	NS	NS

T_1= 0.5 t/ha modified chitosan T_2= 1.0 t/ha modified chitosan

T_3= 2.0 t/ha modified chitosan T_4= 4.0 t/ha modified chitosan

T_5= without modified chitosan

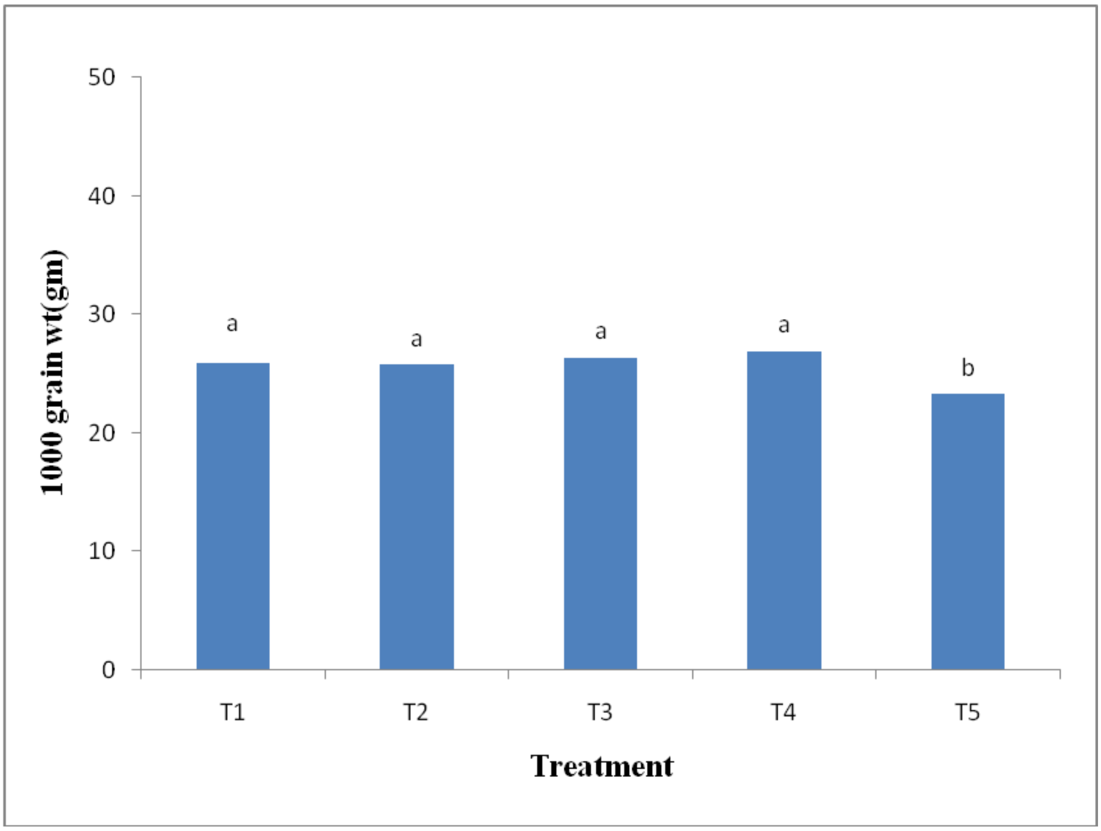


Figure 4. Effects of different doses of modified chitosan on 1000 grain wt. of T. Aman rice (BRRI dhan62). Mean was calculated from three replicates for each treatment. Bars with different letters are significantly different at $p \leq 0.05$ applying DMRT. SAU, 2015.

T_1= 0.5 t/ha modified chitosan T_2= 1.0 t/ha modified chitosan

T_3= 2.0 t/ha modified chitosan T_4= 4.0 t/ha modified hitosan

T_5= without modified chitosan

4.10 pH status of the post-harvest soil

The pH status of the post-harvest soil was affected by the different treatments of modified chitosan and ranged from 5.43 to 6.23 (Fig.5). It was found that pH status of soil was statistically significant. The highest pH value (6.23) was recorded in T_4 treatment (4.0 t/ha modified chitosan). However, the T_4 treatment was significantly greater than from the T_3 (2.0 t/ha modified chitosan), T_2 (1.0 t/ha modified chitosan), T_1 (0.5 t/ha modified chitosan) and T_5 (without modified chitosan) control treatment. The lowest pH value (5.43) was recorded in T_5 treatment (without modified chitosan). According to the pH values treatments may be arranged as $T_4>T_3>T_2>T_1>T_5$. From this study it was observed that, as the rate of modified chitosan application in soil increases the pH status of soil also increases. These results were supported by Kananont *et al*. (2015) who conducted an experiment with Fermented chitin waste (FCW) along with CF = soil supplemented with chemical fertilizer and CMF = soil supplemented with chicken manure fertilizer. The results found that FCW @ 1% the ph differ significantly from 0.5% FCW, 0.25% FCW and the rest of the treatment.

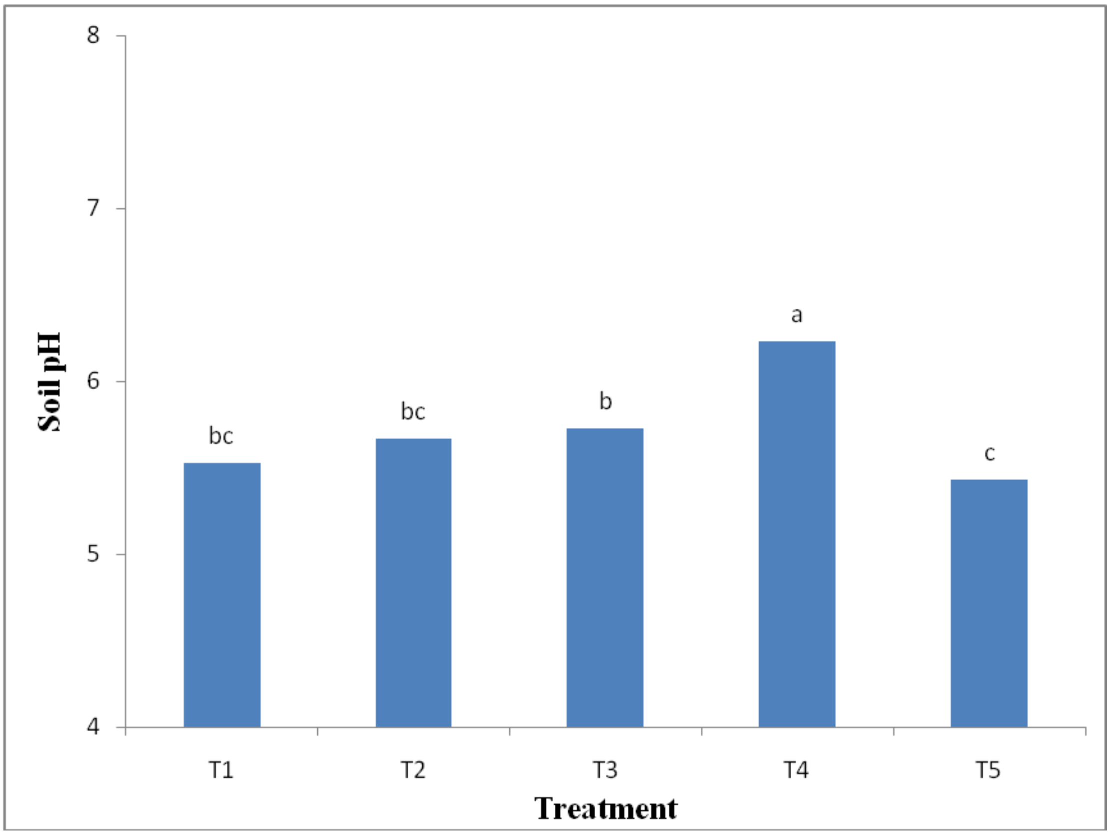


Figure 5. Effects of different doses of modified chitosan on pH status of soil of T. Aman rice (BRRI dhan62). Mean was calculated from three replicates for each treatment. Bars with different letters are significantly different at $p \leq 0.05$ applying DMRT. SAU, 2015.

T_1= 0.5 t/ha modified chitosan T_2= 1.0 t/ha modified chitosan

T_3= 2.0 t/ha modified chitosan T_4= 4.0 t/ha modified chitosan

T_5= without modified chitosan

4.11 Organic carbon content in the post harvest soil

The organic carbon content in the post harvest soil was affected by different treatments of modified chitosan and ranged from 0.63% to 0.97 % (table 4). It was found that Organic carbon content of soil was statistically significant. Maximum organic carbon content (0.97%) was found in T_4 treatment (4.0 t/ha modified chitosan). However, the T_4 treatment was significantly greater than from the T_3 (2.0 t/ha modified chitosan), T_2 (1.0 t/ha modified chitosan), T_1 (0.5 t/ha modified chitosan) and T_5 (without modified chitosan) control treatment. However, minimum organic carbon content (0.63%) was found in T_5 treatment (without modified chitosan). According to the organic carbon content of soil the treatments may be arranged as $T_4>T_3>T_2>T_1>T_5$. From this study it was observed that, as the rate of modified chitosan powder application in soil increases the organic carbon content of soil also increases. These results were supported by Kananont *et al*. (2015) who conducted an experiment with Fermented chitin waste (FCW) along with CF = soil supplemented with chemical fertilizer and CMF = soil supplemented with chicken manure fertilizer. The results found that FCW @ 1% the organic carbon content in soil differ significantly from 0.5% FCW, 0.25% FCW and the rest of the treatment.

The organic carbon content was increased in a dose dependent manner; it might be due to the use of modified chitosan containing higher level of organic carbon level. This result suggests that modified chitosan application might be increase the level of organic matter in soils and would be helpful to improve the sustainable soil health.

4.12 Organic matter content in the post harvest soil

Organic matter content in the post harvest soil showed significant differences among treatments with different modified chitosan doses. Organic matter content ranged from 1.09% to 1.67 % (table 4). Maximum organic matter content (1.67%) was found in T_4 treatment (4.0 t/ha modified chitosan) which was significantly greater than from the T_3 (2.0 t/ha modified chitosan), T_2 (1.0 t/ha modified chitosan) and T_1 (0.5 t/ha modified chitosan) and T_5 (without modified chitosan) control treatment. However, minimum organic matter content (1.09%) was found in T_5 treatment (without modified chitosan). According to the organic matter content of soil the treatments may be arranged as $T_4>T_3>T_2>T_1>T_5$. From this study it was observed that, as the rate of modified chitosan powder application in soil increases the organic matter content of soil also increases. These results were supported by Kananont *et al*. (2015) who conducted an experiment with Fermented chitin waste (FCW) along with CF = soil supplemented with chemical fertilizer and CMF = soil supplemented with chicken manure fertilizer. The results found that FCW @ 1% the organic matter content in soil differ significantly from 0.5% FCW, 0.25% FCW and the rest of the treatment.

The organic matter content was increased in a dose dependent manner, it might be due to the use of modified chitosan containing high amount of organic matter level. Increasing organic matter content for the sustainable agriculture is a big challenge to the Bangladesh soils; however, the modified chitosan application could play a crucial role to increase the organic matter content in soils.

Table 4. Effects of different doses of modified chitosan on soil organic status of T. Aman rice (BRRI dhan62) at harvest. SAU, 2015.

Treatment (dose)	Soil organic carbon (%)	Soil organic matter (%)
T_1	0.72d	1.25d
T_2	0.81c	1.40c
T_3	0.89b	1.53b
T_4	0.97a	1.67a
T_5	0.63e	1.09e
LSD (0.05)	0.05	0.09
CV (%)	3.50	3.60
Level of significance	**	**

Values in a column with different letters are significantly different at $p \leq 0.05$ applying LSD.

** = Significant at 1% level of probability

T_1= 0.5 t/ha modified chitosan
T_2= 1.0 t/ha modified chitosan
T_3= 2.0 t/ha modified chitosan
T_4= 4.0 t/ha modified chitosan
T_5= without modified chitosan

4.13 Grain yield (t/ha)

Figure 6 shows the effects of different treatments on grain yield. Grain yield was significantly influenced by the modified chitosan treatment. Grain yield was increased due to the modified chitosan treatment compare to the control treatment. The highest grain yield (6.15 t/ha) was obtained in the T_4 (4.0 t/ha modified chitosan) treatment which was significantly greater than that obtained in the T_5 control (without modified chitosan) and T_1 treatment (0.5 t/ha modified chitosan) and statistically identical to T_2 (1.0 t/ha modified chitosan) and T_3 (2.0 t/ha modified chitosan) treatment. However the grain yield did not differ significantly in T_2, T_3, T_4 treatments. The lowest grain yield (4.49 t/ha) was obtained in the T_5 (without modified chitosan) control treatment which is statistically identical to T_1 treatment. In terms of grain yield the treatments may be arranged as $T_4 > T_3 > T_2 > T_1 > T_5$. It was observed that, as the rate of modified chitosan application in soil increases grain yield also increases. These results were supported by Sultana *et al*. (2015) who conducted a field experiment with Four different concentrations that is 0, 40, 80 and 100 ppm oligomeric chitosan and four times foliar spray after germination were carried out. Finally it is observed that grain yield of rice show significant differences between control plants and foliar sprayed chitosan plants. Similar results were also found by Boonlertnirun *et al.* (2006), Boonlertnirun *et al*. (2007), Boonlertnirun *et al*. (2008) and Kananont *et al*. (2015).

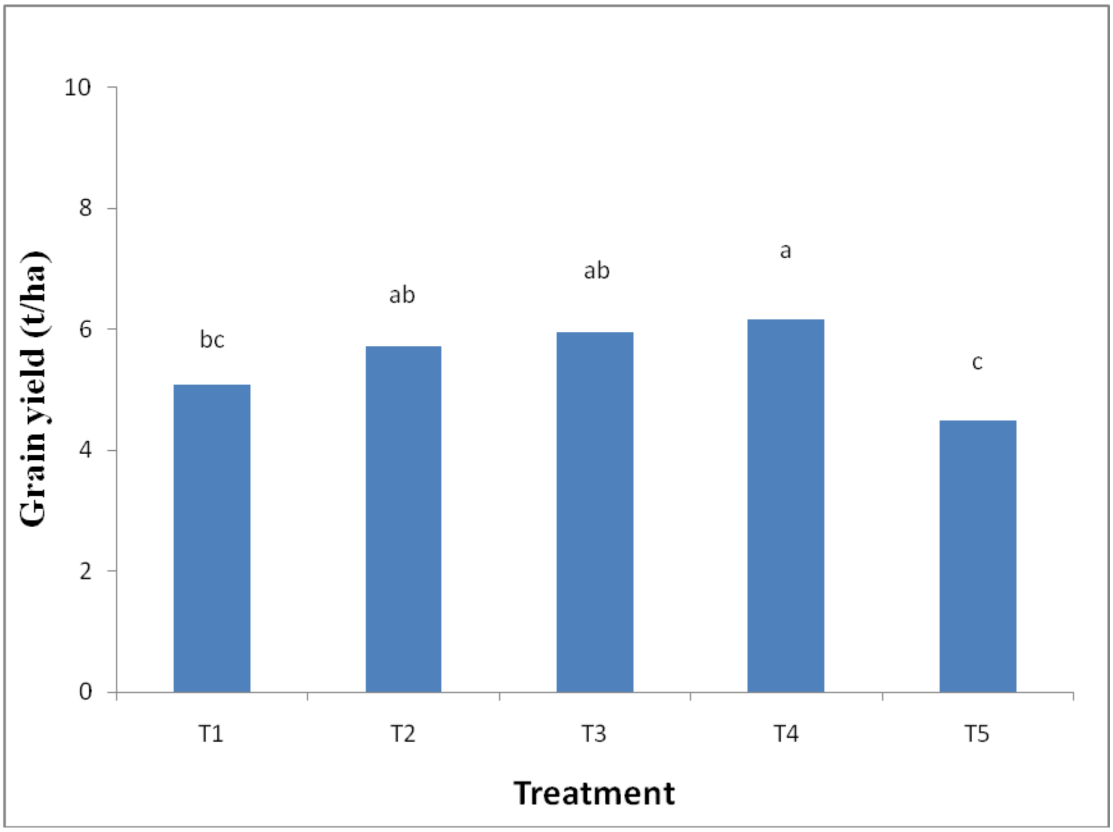


Figure 6. Effects of different doses of modified chitosan on grain yield of T. Aman rice (BRRI dhan 62). Mean was calculated from three replicates for each treatment. Bars with different letters are significantly different at $p \leq 0.05$ applying DMRT. SAU, 2015.

T_1= 0.5 t/ha modified chitosan

T_2= 1.0 t/ha modified chitosan

T_3= 2.0 t/ha modified chitosan

T_4= 4.0 t/ha modified chitosan

T_5= without modified chitosan

4.14 Straw yield (t/ha)

The effects of different treatments on straw yield were significantly influenced by the modified chitosan treatment (Fig 7). Straw yield was increased due to the modified chitosan treatment compare to the control treatment. The highest straw yield (6.80 t/ha) was obtained in the T_4 (4.0 t/ha modified chitosan) treatment which was significantly greater than that obtained in the T_5 control (without modified chitosan) and T_1 treatment (0.5 t/ha modified chitosan) and statistically identical to T_2 (1.0 t/ha modified chitosan) and T_3 (2.0 t/ha modified chitosan) treatment. However the straw yield did not differ significantly in T_2, T_3, T_4 treatments. The lowest straw yield (5.07 t/ha) was obtained in the T_5 (without modified chitosan) control treatment which is statistically identical to T_1 treatment. In terms of straw yield the treatments may be arranged as $T_4>T_3>T_2>T_1>T_5$. It was observed that, as the rate of modified chitosan application in soil increases straw yield also increases. These results were supported by Sultana *et al*. (2015) who conducted a field experiment with Four different concentrations that is 0, 40, 80 and 100 ppm oligomeric chitosan and four times foliar spray after germination were carried out. Finally it is observed that straw yield of rice show significant differences between control plants and foliar sprayed chitosan plants.

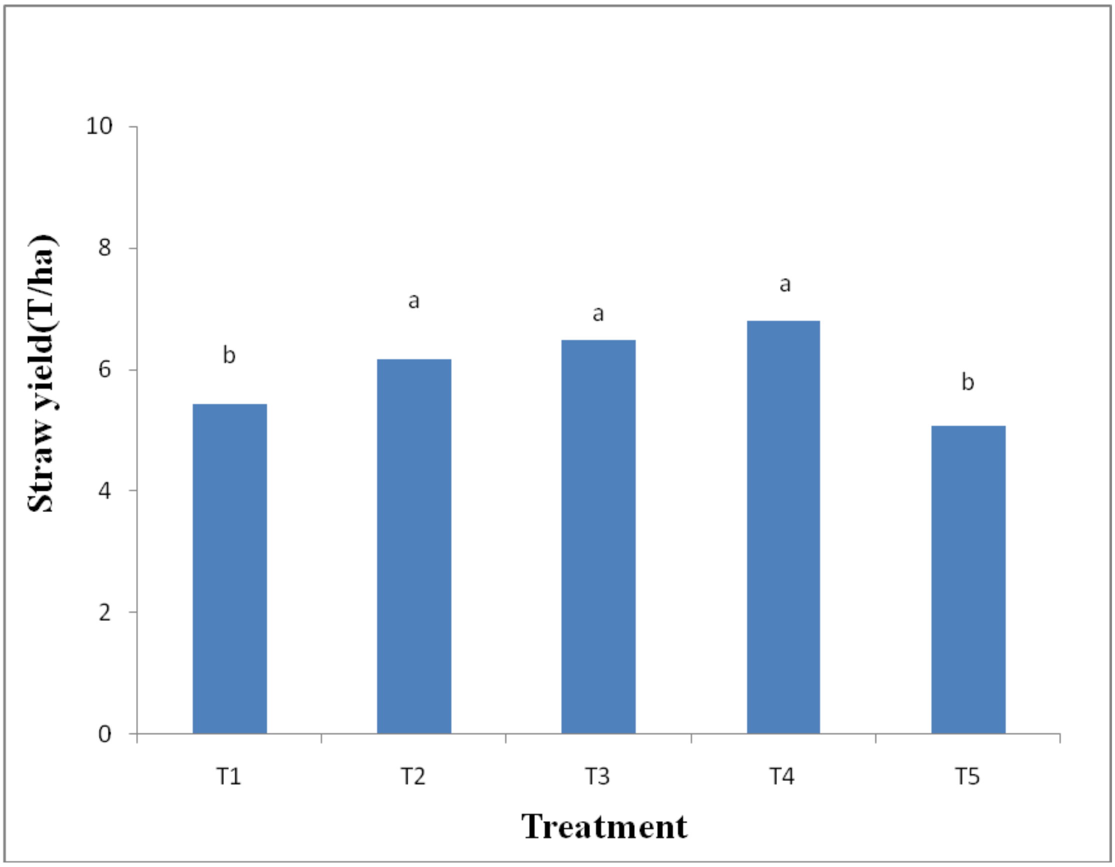


Figure 7. Effects of different doses of modified chitosan on straw yield of T. Aman rice (BRRI dhan 62). Mean was calculated from three replicates for each treatment. Bars with different letters are significantly different at *p* ≤ 0.05 applying DMRT. SAU, 2015.

T_1= 0.5 t/ha modified chitosan

T_2= 1.0 t/ha modified chitosan

T_3= 2.0 t/ha modified chitosan

T_4= 4.0 t/ha modified chitosan

T_5= without modified chitosan

4.15 Biological yield (t/ha)

Biological yield were significantly influenced by the modified chitosan treatment (Table 5). Biological yield was increased due to the modified chitosan treatment compare to the control treatment. The highest biological yield (12.95 t/ha) was obtained in the T_4 (4.0 t/ha modified chitosan) treatment which was significantly greater than that obtained in the T_5 control (without modified chitosan) and T_1 treatment (0.5 t/ha modified chitosan) and statistically identical to T_2 (1.0 t/ha modified chitosan) and T_3 (2.0 t/ha modified chitosan) treatment. However the biological yield did not differ significantly in T_2, T_3, T_4 treatments. The lowest biological yield (9.560 t/ha) was obtained in the T_5 (without modified chitosan) control treatment which is statistically identical to T_1 treatment. In terms of biological yield the treatments may be arranged as $T_4>T_3>T_2>T_1>T_5$. It was observed that, as the rate of modified chitosan application in soil increases biological yield also increases.

4.16 Harvest Index

Harvest index (HI) is the ratio of seed yield to total above ground plant yield. Significant response was not observed in the harvest index due to the effect of different modified chitosan treatments on BRRI dhan62 (Table 5). From the results, it was found that the highest harvest index (48.39%) was obtained from the treatment T_1 (0.5 t/ha modified chitosan) and the lowest harvest index (46.91%) was obtained in the T_5 control treatment (without modified chitosan)

Table 5. Effects of different doses of modified chitosan on Biological yield and harvest index of T. Aman rice (BRRI dhan62) at harvest. SAU, 2015.

Treatments	Grain yield (t/ha)	Straw yield (t/ha)	Biological yield (t/ha)	Harvest index (%)
T_1	5.09bc	5.43b	10.51b	48.39
T_2	5.71ab	6.16a	11.87a	48.16
T_3	5.94ab	6.49a	12.43a	47.68
T_4	6.15a	6.80a	12.95a	47.37
T_5	4.49c	5.07b	9.56b	46.91
LSD (0.05)	0.97	0.66	1.23	5.15
CV (%)	9.46	5.86	5.68	5.74
Level of significance	*	**	**	NS

Values in a column with different letters are significantly different at $p \leq 0.05$ applying LSD.

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

T_1= 0.5 t/ha modified Chitosan

T_2= 1.0 t/ha modified Chitosan

T_3= 2.0 t/ha modified Chitosan

T_4= 4.0 t/ha modified Chitosan

T_5= without modified Chitosan

Table 6. Composition of the modified chitosan

Name of the nutrients	Nutrient content
Nitrogen (N)	4.06 %
Phosphorus (P)	0.643 %
Potassium (K)	0.28 %
Sulphur (S)	0.092 %
Calcium (Ca)	2.43 %
Magnesium (Mg)	0.36 %
Zinc (Zn)	92.03 ppm
Boron(B)	152 ppm
Organic Carbon (OC)	7.52 %
Organic Matter (OM)	12.96 %
pH of the Modified Chitosan	8.73

Analytical results (Table 6) revealed that a number of essential (macro and micro elements) were supplied due to the application of the modified chitosan in the rice field soils. With the alkaline behavior of the materials increased the pH level of the rice field soils. Many factors could be involved in the supper growth, development and yield increment of the rice grain. The above mentioned nutritional supplementation and some other growth promoting hormone could be involved in the mechanisms.

CHAPTER V

SUMMARY AND CONCLUSION

An experiment was carried out at the research field of the Department of Soil science, of Sher-e-Bangla Agricultural University, Dhaka-1207 during the period from July to December 2015. The study was aimed to determine the effect of modified chitosan on growth, yield contributing characters and yield of rice cv. BRRI dhan62, a short duration, zinc enriched, and T. Aman variety. The experiment comprised of five levels of modified chitosan treatments and was: T_1 (0.5 t/ha modified chitosan), T_2 (1.0 t/ha modified chitosan), T_3 (2.0 t/ha modified chitosan), T_4 (4.0 t/ha modified chitosan) and T_5 (without modified chitosan). The single factor experiment was laid out in a randomized complete block design (RCBD) with three replications. Modified chitosan was applied and mixed in the soil before transplanting of rice. Modified chitosan effect data were recorded on plant height (cm) at harvest, number of total tillers/hill, number of effective tillers/hill, panicle length, number of filled grains/panicle, unfilled grains/panicle, 1000 grain weight, pH, grain yield, straw yield, biological yield and harvest index.

Application of modified chitosan had a profound influence on morphological, reproductive, yield attributes and grain yield of rice. Modified chitosan also improves and increase pH, organic carbon and organic matter content of soil. Among the modified chitosan concentrations, 4.0 t/ha modified chitosan perform best among the significantly varied parameters compare to control and other concentrations of chitosan. But in case of non significant parameters the treatment may differ.

The results revealed that the maximum total tillers/hill (18.83) was observed in the T_4 (4.0 t/ha modified chitosan) treatment and minimum total tillers/hill (15.28) was observed in the T_5 (without modified chitosan) control treatment. Similarly highest effective tillers/hill (17.85) was observed in the T_4 (4.0 t/ha modified chitosan) treatment and lowest effective tillers/hill (15.09) was observed in the T_5 (without modified chitosan) control treatment. Similarly highest grain yield (6.15 t/ha), highest straw yield (6.80 t/ha), highest biological yield (12.95 t/ha), maximum 1000 grain weight (26.90gm), highest value of pH (6.23), highest organic carbon content (0.97%), highest organic matter content (1.67%) was observed in the T_4 (4.0 t/ha modified chitosan) treatment and lowest grain yield (4.49 t/ha), lowest straw yield (5.07 t/ha), lowest biological yield (9.56 t/ha), minimum 1000 grain weight (23.27gm), lowest value of pH (5.43), lowest organic carbon content (0.63%), lowest organic matter content (1.09%) was observed in the T_5 (without modified chitosan) control treatment.

Application of modified chitosan did not affect plant height, panicle length, total number of grains/panicle, filled grains/panicle, unfilled grains/panicle but the grain number increased with the increasing doses of modified chitosan among the treatments compare to the control. Based on the experimental results, it might be concluded that,

i. modified chitosan influenced morphological characters, yield attributes and grain yield of rice cv. BRRI dhan62 over control; and

ii. all the treatments (T_1, T_2, T_3, T_4) using modified chitosan showed better performance than control (T_5) and the performance of the treatments (T_2, T_3, T_4) were similar but the treatment T_4 was the superior than all the treatments.

iii. The modified chitosan improved chemical properties of soil for sustainable agriculture.

Recommendations

From the above experimental findings, it is apparent that the application of modified chitosan @ 4.0 t/ha (T_4) performed better on yield and yield parameters of rice cv. BRRI dhan62. In order to recommend the practices for the rice growers, the following aspects would be considered in future:

i) Similar experiments need to be conducted in different locations and seasons of Bangladesh to draw a final conclusion regarding the modified chitosan applications for the grain yield of rice.

ii) Varietal trials need to be investigated.

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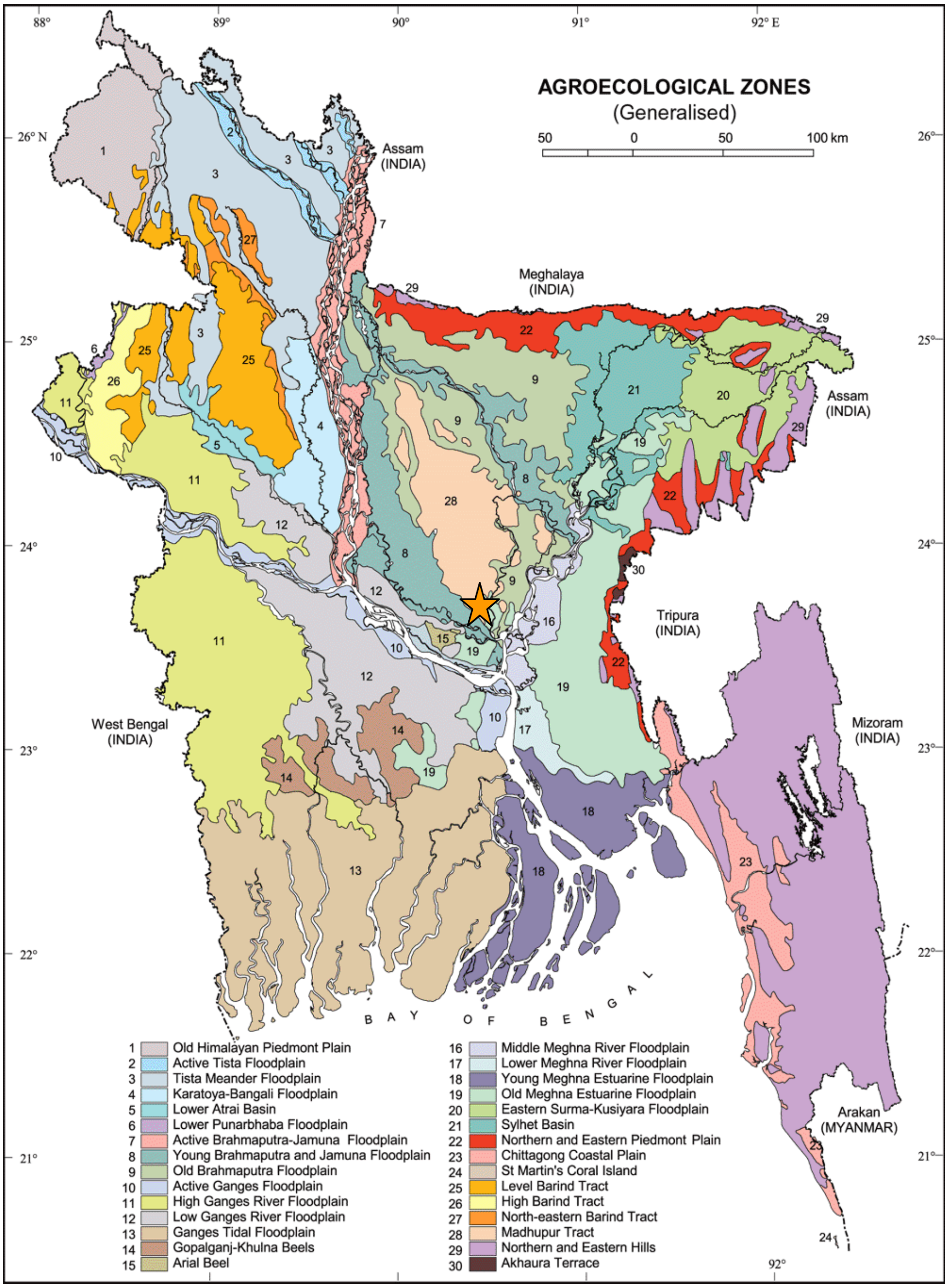
APPENDICES

Appendix I. Map showing the experimental sites under study



Appendix II. Monthly record of air temperature, relative humidity, rainfall and sunshine (average) of the experimental site during the period from July to December 2015, SAU.

Month (2016)	Air temperature(^{0}C)		Relative humidity (%)	Rainfall (mm)	Sunshine (hr)
	Maximum	Minimum			
July	35.5	23.2	78	312	5.4
August	36.0	24.5	83	563	5.1
September	36.0	23.5	81	319	5.0
October	34.5	24.4	81	279	4.4
November	26.5	19.4	81	22	6.9
December	25.8	16.0	78	00	6.8

Source: Bangladesh Meteorological Department (Climate and weather division) Agargoan, Dhaka-1212

Appendix III. Analysis of variance (mean square) of morphological, soil parameters and yield components of T. Aman rice (cv. BRRI dhan62) SAU, 2015.

Source	DF	Plant height	Total tiller	Effective tiller	Un-effective tiller	Panicle length	Total grain	Filled grain	Unfilled grain	Grain yield	Staw yield	Biological yield	Harvest index	1000 grain wt	pH	OC%	OM%
Rep	2	7.2797	0.22965	0.57498	0.10891	1.0905	73.862	37.962	12.248	0.50405	0.14435	1.12358	2.56189	0.13067	0.006	0.00002	0.00003
Treat	4	2.8612NS	8.13781**	5.48927**	0.29063NS	0.58237NS	52.529NS	50.6717NS	4.4107NS	1.38233*	1.58117**	5.88324**	1.06951NS	5.78433**	0.30167**	0.5299**	0.15711**
Error	8	11.8547	0.29395	0.5568	0.07947	0.67307	106.225	71.4012	17.4547	0.26851	0.12301	0.42468	7.49155	0.56233	0.01267	0.0008	0.0025

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

DF: Degrees of freedom

OC: Organic carbon

OM: Organic matter