# **Application of Chitosan in Rice Production**

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#### Abstract

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Chitosan is a natural biopolymer which stimulates growth and increases yield of plants as well as induces the immune system of plants. The objectives of this study were to determine appropriate applications for increasing of the rice yield. The experiment was performed with randomized complete block design (RCBD) with four replications. Four treatments were done as follows: no chitosan application, seed soaking with chitosan solution, seed soaking and soil application with chitosan solution and seed soaking and foliar spraying with chitosan solution. The results showed that application of chitosan by seed soaking and soil application four times throughout cropping season significantly increased rice yield over the other treatments whereas application by seed soaking and spraying the foliar four times tended to show an ability on disease control. However, it did not show statistically significant differences when compared with the control.

**Key words**: chitosan application, rice production

## Introduction

Chitosan is a natural biopolymer modified from chitin, which is the main structural component of squid pens, cell walls of some fungi and shrimp and crab shells. Chitin and chitosan are copolymers found together in nature. They are inherent to have specific properties of being environmentally friendly and easily degradable. Thailand is a world-leading exporter of frozen shrimps. Therefore, there are abundant raw materials for chitosan production. Chitosan has a wide scope of application. With high affinity and non-toxicity, it does no harm 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. (5) Chitosan has strong effects on agriculture such as acting as the carbon source for microbes in the soil, accelerating of transformation the process of organic matter into inorganic matter and assisting the root system of plants to absorb more nutrient from the soil. Chitosan is absorbed to the root after being decomposed by bacteria in the soil. 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. (14, 3)

## **Materials and Experimental Procedures**

The experiment was done with randomized complete block with four replications. The treatments were Tr 1- no chitosan application (control), Tr 2- seed soaking with chitosan solution, Tr 3 - seed soaking and soil application with chitosan Tr4 - seed soaking and foliar solution and spraying with chitosan solution. One experimental unit comprised six pots. Rice seeds cv. Suphanburi 3 were soaked in polymeric chitosan solution at the concentrate of 80 ppm for 4-5 hours before planting for Tr2, Tr3 and Tr4 whereas rice seeds in the control treatment were soaked in distilled water. Eight rice seeds were planted in an 18 inchdiameter pot containing paddy soil. When rice seeds grew up to be seedlings they were thinned out to allow four healthy plants per pot for data recording. When the age of rice plants was 14, 24, 34 and 44 days after planting so as chitosan at the concentration of 80 ppm was applied by soil application and foliar spraying for Tr 3 and Tr4 respectively. Chemical fertilizer mixed between

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urea(46-0-0) and paddy field fertilizer(16-20-0) (ratio:1:1w/w) was also applied at the rate of 50 kg/rai (~20 g/pot) in all treatments. Plant height, leaf greenness, tiller numbers, yield and yield components were recorded at harvesting time. The infected leaves were recorded after inoculation for ten days. Fiber percentage was assayed when rice plants completely bloomed (heading stage). All data were subjected to analysis of variance according to the experimental design used in this study and Least Significant Difference (LSD) was utilized to compare the different means of treatment.

#### **Results and Discussion**

#### Growth

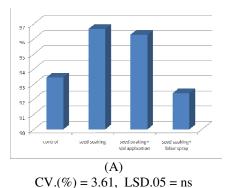
## Plant Height

Rice plants were not influenced by varying methods of application. Seed soaking in chitosan solution before planting tended to stimulate plant height. However, it did not show any statistically significant differences from the others, Figure 1(A). This result was supported by the work of who found that foliar application of oligomeric chitosan did not affect soybean height. On the other hand, 13 revealed that chinese cabbage seeds incorporated with chitosan at the rate of 0.4-0.6 mg/g of seed followed by foliar spraying at 20-40 mg/l reacted with increased plant height and leaf area of chinese cabbage plants.

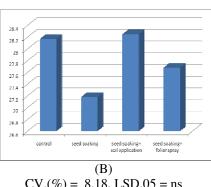
## Leaf Greenness

Leaf greenness of rice plants measured by chlorophyll meter (SPAD 502) was not significantly different from treatments that applied chitosan with different application methods. However, soaking the seeds in chitosan solution before planting and soil application four times tended to show the maximum value of leaf greenness over the other treatments, Figure 1(B). Yue, *et al.* 2001 found that the changing regulation of chlorophyll content (leaf greenness) correlated with the change in chitosan concentration. Moreover, reported that tomato and lecttuce leaves turned darker green with increasing chitosan concentration.

## Rice height (cm)



## Leaf greenness (spad unit)



CV.(%) = 8.18, LSD.05 = ns

**Figure 1.** Effect of various chitosan application methods on rice height (A) and leaf greenness (B) of rice plants.

## Tiller Numbers Per Plant

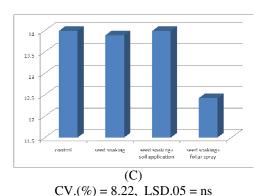
Varying chitosan application methods did not affect tiller numbers per plant. The maximum tiller numbers obtained from treatment of seed soaking in chitosan solution before planting and soil application, however did not significantly differ from the control, Figure 2(C). A similar result of showed that node and branch numbers of soybean increased after application of chitosan in the soil. Ohta, *et al.* 2001 also reported that the application of soil mixed with chitosan 1%w/w at sowing remarkably increased flower numbers of *Eustoma grandiflorum*.

#### Dry Matter Accumulation

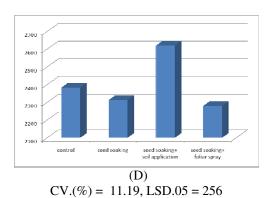
Dry matter accumulation of rice plants showed significant differences among various application methods. Soaking the seeds in chitosan

solution before planting and application in soil four times gave the maximum dry matter (2620 g/Tr.), whereas the control treatment gave only 2,383 g/Tr. Dry matter accumulation gained from treatment with seed soaking and foliar spraying was the lowest (2,280g/Tr.), Figure2(D). This result was supported by<sup>(4)</sup> Chibu and Shibayama (1999) who found that dry weight of dry-land rice cv. Misatohatamochi grown with both 0.1 and 0.5% of chitosan were increased over the control. In addition,<sup>(7)</sup> indicated that tomato plants grown from seed coated with chitosan showed greater dry weight and stem thickness than untreated tomato plants.

#### Tiller no./plant (tiller)



Dry matter/Tr\* (g)



**Figure 2.** Effect of various chitosan application methods on tiller numbers (C) and dry matter (D) of rice plants.

\* Tr = comprised 6 pots (24 plants/Tr)

## Yield and Yield Components

#### Grain Yield

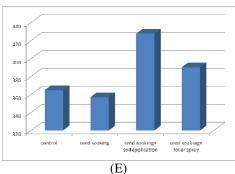
The maximum yield has achieved by applying chitosan by seed soaking and soil application; the

was 427.78 g/Tr, whereas average yield control was 364.83 g/Tr. The statistically significant differences were detected among Figure 3(F). This result could explain that application of chitosan by incorporation in soil showed the maximum grain yield since chitosan- availability periods in soil were longer than those of foliar spraying. Moreover, chitosan had a positive ionic charge which chemically bind with plant nutrients that showed a negative ionic charge resulting in a slowly released action in rice plants which closely contributed to yield increasing. There were many reports supporting this result such as the report of (8) who found that panicle numbers and yield of wheat (Triticum aestivum L.) were increased after application of polymeric or oligomeric chitosan. Boonlertnirun, et al. 2006 reported that rice yield cultivar Suphan Buri 1 was significantly increased over the control (no chitosan) after application of polymeric chitosan at the concentration of 20 ppm.

#### Panicle Numbers Per Plant

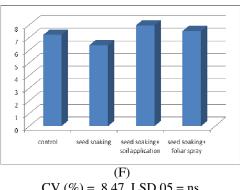
The issue of panicle numbers is one of the important yield components which directly affected rice yield. No statistically significant differences were found among chitosan application with different methods including the control. The maximum number of panicles has obtained from treatment with seed soaking before planting and soil application later, whereas the treatment with seed soaking in chitosan solution showed the minimum number of panicles, Figure3(F). Lu, *et al.* 2002 found that the panicle numbers of rice were increased after watering with chitosan at the rate of 0.4 g/50 cm³ (chitosan: water).

## Grain yield/Tr\* (g)



CV.(%) = 15.05, LSD.05 = 52.13

# Panicle no./plant(panicle)



CV.(%) = 8.47, LSD.05 = ns

Figure 3. Effect of various chitosan application methods on grain yield (E) and panicle numbers (F) of rice

\* Tr = comprised 6 pots (24 plants/Tr)

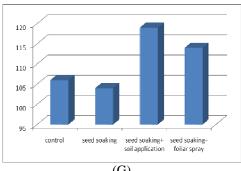
#### Seed Numbers Per Panicle

Seed numbers per panicle were not influenced by various chitosan application methods. Application of chitosan by seed soaking in chitosan solution before planting and then applying in soil tended to produce more panicle numbers than the other methods. However, it was not significantly different from the other treatments and the control. The lower number of panicles was found in treatment of seed soaking in chitosan solution, Figure 4(G). This result was supported by the work of<sup>(1)</sup> who indicated that seed numbers per panicle of rice plant cv. Suphan Buri 1 were not affected by various chitosan concentrations.

## 1,000-Grain Weight

Application of chitosan by varying application methods did not affect 1,000- grain weight. The maximum seed weight was gained from seed soaking in chitosan solution before planting and then applying in soil whereas chitosan application by seed soaking in chitosan solution before planting and then foliar spraying showed the minimum seed weight. Nevertheless, no significant difference was found among treatments, Figure 4(H). This was contrary to the observations of who found that thousand grain weight of wheat plants was increased with application of polymeric chitosan at low concentration.

## Seed no./panicle (seed)



(G)CV.(%) = 12.11, LSD.05 = ns

## 1,000-grain weight(g)

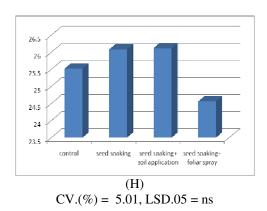


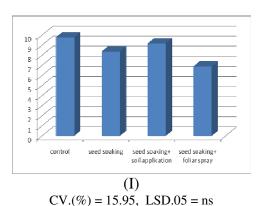
Figure 4. Effect of various chitosan application methods on seed numbers(G) and 1,000-grain weight (H) of rice plants.

## Infected Leaves and Fiber Percentage

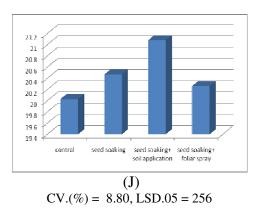
The infected rice leaves observed in treatment of seed soaking in chitosan solution before planting and foliar application later were rather slight; the percentage was 6. 9 %, whereas infected leaves in the control amounted to 9.78 %, Figure 5(I). There were many academic papers reporting on chitosan acting as elicitor such as the work of (10) which revealed that the plants with high content of chitin enzyme had better external disease resistance to pathogen than the others. Moreover, chitosan was shown to be able to activate plant defensive genes through the octadecanoid pathway. (5) Considering fiber percentage, application of chitosan by seed soaking before planting and four times of soil application throughout cropping season increased fiber percentage in rice plants greater than the other treatments. However, no statistically significant difference was found, Figure 5(J). An explanation may be the following:

if rice leaves contain high fiber percentage, there is a tendency to produce an ability to protect disease and insect infection which is greater than with those of low fiber percentage due to hard infection.

## Infected leaves(%)



fiber percentage(%)



**Figure 5.** Effect of various chitoson application methods on infected leaves (I) and fiber percentage (J) of rice plants.

## **Conclusions**

This study implies that application of 80 ppm polymeric chitosan by seed soaking before planting followed by soil application for four times throughout cropping season tended to stimulate growth and significantly increased rice yield. On the other hand, seed soaking before planting followed by foliar application tended to generate an ability in disease control.

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