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Indian J.Sci.Res. 15 (1): 31-38, 2024 **Original Research Article**

CHARACTERIZATION OF LATE SOWING BORO SEEDLINGS BY USING CHITOSAN RAW MATERIAL POWDER

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ABSTRACT

An experiment was conducted inside the net house of Sher-e-Bangla Agricultural University (SAU), Dhaka, Bangladesh, from November 2019 to April 2020, to investigate the improvement of late sowing *Boro* seedling characters by using chitosan raw material powder in seedbed soil. The experiment followed Completely Randomized Design (CRD) with four replications. Level of chitosan raw material powder(w/w) (06) *viz;* C_0= 0%, C_1= 0.1%, C_2= 0.2%, C_3= 0.3%, C_4= 0.4% and C_5= 0.5%. The study found that the application of chitosan raw material powder significantly improved seedling growth and reduced fungal infection risk than untreated seedlings. Experimental results revealed that among different levels of chitosan raw material powder, seedlings treated with C_3 (0.3 %) treatment perform well and recorded the maximum average seedling height (17.75 cm), fresh weight seedling^{-1} (531.67 mg), oven dry weight seedling^{-1} (96.67 mg) & seedling strength (5.44 mg/cm). In case of seed bed soil, maximum P^H, percent organic carbon & percent organic matter was observed in C_5 (0.5 % Chitosan) treatment that was 6.50, 0.76 & 1.31 respectively but maximum nitrogen percent was recorded in C_4 (0.4 % chitosan) treatment that was 0.12. Chitosan raw material powder increases soil microbial activity, acts as a plant growth regulator, increases soil resource and water holding capacity as well as helps other parameters of soil nutrients. In this research work, we examine the potential of chitosan raw material powder as a novel intervention to improve the performance of late-sown Boro rice seedlings.

KEYWORDS: Chitosan, Late Sowing, Rice, Seedling, Soil, and Seedling Strength

Rice (*Oryza sativa* L.) is the most important food crop and a primary food source for more than one-third of the world's population (Sarkar *et al.*, 2017). Maclean et al., 2002 projected a 25% increase in the world's rice demand from 2001 to 2025 to keep pace with population growth. Bangladesh faces challenges in meeting this demand sustainably as natural resources shrink and rice significantly impacts the country's agrarian economy, with annual production reaching 36.28 million tons. (BBS, 2018). Bangladesh's rice production is predicted to reach 36.3 million tons in 2021 due to hybrid and high-yield variety cultivation, and imports are expected to ease COVID-19 food security concerns. (USDA, 2021). Three distinct seasons primarily produce Bangladesh's rice production: Boro (55.50%) (December-May), Aman (37.90%) (July-November), and Aus (6.60%) (April-June), with Boro accounting for over half of the total production. (APCAS, 2016). Among three growing seasons (Aus, Aman and Boro), *Boro* rice is the most important rice crops for Bangladesh with respect to its high yield and contribution to rice production. *Boro* cultivation area has declined to 4.75 million hectares in 2020, which was 4.9 million hectares in 2019. The country produced an all-time-high 20.03 million tons of *Boro* rice in 2019. The government expects to achieve 20.04 million tons of *Boro* production target but many farmers are upset with low paddy and rice prices, switched to other crops like corn, vegetables and tobacco etc. (Express, 2021). Bangladesh's food security is a pressing issue, particularly in rice production, with an average yield of less than 50% of the world average. In Bangladesh, the national mean yield of rice (2.60 t ha-1) is lower than the potential national yield (5.40 t ha-1) and world average yield (3.70 t ha-1) (Pingalie et al., 1997). Adopting modern cultivars, implementing nutrient management practices, and utilizing advanced technology such as seedling age can improve the lower yield of transplanted Boro rice.

Chitosan is a natural polymer and one of the chitin derivatives when the degree of deacetylation of chitin reaches about 50% (Rinaudo, 2006). Deacetylation of chitin forms Chitosan, a linear polymer with two subunits, D-glucosamine and N-Acetyl-D-glucosamine, linked by glycosidic bonds. (Hidangmayum *et al.*, 2019). Molecular weight and the degree of deacetylation influence the

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functional properties of chitosan, including its solubility, biodegradability, and diverse bioactive attributes (Rajoka et al., 2019). Many studies have differently determined classes of chitosan based on its molecular weight; however, the specific categories are still unclear. Commercially, we classify chitosan into three main classes: low (50–190 kDa), medium (190-310 kDa), and high (310–375 kDa) molecular weight (MW). (Prashanth and Tharanathan, 2007*).* Chitosan is different from most neutral or negatively charged polysaccharides because it is cationic. This means that it can form electrostatic complexes with other naturally or synthetically negatively charged polymers. (Rinaudo, 2006). Therefore, researchers have investigated and developed chitosan as a plant biostimulant (Katiyar et al., 2015; Hidangmayum et al., 2019). Chitosan, a plant defense response, effectively increases plant productivity and protects against pathogen attacks, utilizing its defense responses in various plant-related applications. (Malerba and Cerana, 2018). Previous studies revealed that chitosan has the potential to enhance plant growth as well as increase yield in many crops, including apple, wheat, maize, and rice (Yang *et al*., 2009; Lizárraga-Paulín *et al*., 2011; Zeng and Luo, 2012; Seang-Ngam *et al*., 2014)

MATERIALS AND METHODS

The study was taken at Sher-e-Bangla Agricultural University, Dhaka, aimed to improve the late-sowing Boro seedling characteristics by using Chitosan raw material powder. Various adversities constantly confront farmers in our country during their cultivation process. Some areas produce crops later than the fixed season period. Particularly in areas that produce perennial crops, there is a slight delay in reaching the final cultivation period of two crops, which in turn leads to the production of the next crop. The production is slightly delayed. In this situation, farmers often face challenges due to weather or climate temperature variations. This disrupts production, leading to food shortages which lead economic losses. We apply chitosan to the soil to minimize this problem, thereby increasing crop production. Farmers may choose to use chitosan raw powder for late-sown boro seedlings to enhance their growth and resilience. Chitosan has shown promise in promoting root development, improving nutrient uptake, and increasing tolerance to environmental stressors. Late-sown boro seedlings face challenges such as shorter growing periods and exposure to adverse weather conditions, making them more vulnerable to stress and disease. The experiment was conducted from November 2019 to April 2020 in the agronomy field at 23°77′ N latitude and 90°33′ E longitude. (Anon., 2004). The experimental site is in the agro-ecological zone of "The Modhupur Tract," AEZ-28. The soil is shallow red brown terrace soil, with pH ranged 5.4-5.8, and is above normal flood level. Soil samples were collected from Sher-e-Bangla Agricultural University Farm and analyzed them at the Soil Resource and Development Institute, Dhaka. Physio-chemical properties of the collected soil were shown at table 1.

Table 1: The initial physical and chemical characteristics of soil used in this experiment

Constituents	Sand	silt	clay	Textual class	Soil	pH	Organic carbon (%)	Organic matter (%)	Total nitrogen (%)	Available P (ppm)	Exchangeable K (mg/100 g soil)
%	26	45	29	Silty clay	Value	5.8	0.5	.87	.04	20.54	.10

Experimental Materials

BRRI dhan88 and different level of chitosan raw material powder were used as experimental material for this experiment, focusing on the popular BRRI dhan88, which was collected from Bangladesh Rice Research Institute (BRRI), Gazipur. BRRI dhan88 seeds were chosen for their health and disease-free nature. After 24 hours of immersion in water, then stored in gunny bags, sprouting after 48 hours and ready for sowing 72 hours late. Seeds contain 76% carbohydrate, 26.3% amylose and 9.8% protein.

Seed pot preparation, application of chitosan raw material powder and Experimental treatment

Seedlings were grown using 2-inch plastic pots, moist soil from Sher-e-Bangla Agricultural University farm, mixed with different levels of Chitosan raw material powder according to par treatment requirements. Then the pot was filled with 1 kg of chitosan raw material-treated soil. After that 100 seeds were sown in the pot for raising seedlings.

Level of Chitosan raw material powder (w/w) (6) *viz;*

C_0= 0%, C_1= 0.1%, C_2= 0.2%, C_3= 0.3%, C_4= 0.4% & C_5= 0.5%

Table 2: Composition of Chitosan powder

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%

The Procedure for Data Collection

We recorded the data using the following parameters:

1. Average seedling height (cm)
2. Fresh-weight seedling (1 g)
3. Oven dry weight seedling^{-1} (g)
4. Seedling strength (mg/cm)

Using a meter scale, the study measured the height of 25 seedlings during transplanting and expressed the mean height in cm. We collected and weighed the fresh weight of the 25 seedlings using a digital electric balance. We collected 25 differently treated, sun-dried seedlings, dried them in an oven, and expressed their mean weight in mg. We measured the seedling strength using the following formula:

$$\text{Seedling strength}=\frac{Oven\ dry\ weight\ per\ seedling}{Average\ seedling\ height}\ mg/cm$$

We conducted a chemical analysis of the soil in the seed pot after transplanting the seedlings.

Particle Size Analysis and pH

We conducted a soil particle size analysis using the hydrometer method, determined the textural class using the sand, silt, and clay values in the USDA's "MarshalL-1s Textural Triangular Coordinate," and measured the soil pH using a glass electrode pH meter, using soil and water at a ratio of 1:2.5, as described by Jackson (1962).

Organic C

We used the Walkley and Black's Wet Oxidation Method to determine the organic carbon in soil, oxidizing it with 1N $K_2Cr_2O_7$ in H_2SO_4 and titrating it with 1N $FeSO_4$.

Total Nitrogen

We used the MicroKjeldahl method to determine the soil nitrogen content. We heated and digested a soil sample before titrating it with standard 0.01 N H_2SO_4. The nitrogen content was calculated using a formula: % N = (T-B) × N × 0.014 × 100/S, where T represents the sample titration value, B represents the blank titration value, N represents the strength of H_2SO_4, and S represents the sample weight.

Data Analysis Technique

A computer package program named MSTAT-C assisted in compiling and statistically analyzing the collected data using the analysis of variance (ANOVA) technique. Duncan's Multiple Range Test (DMRT) estimated the significance of the difference among the treatment means at the 5% level of probability (Gomez and Gomez, 1984)

RESULTS AND DISCUSSION

Effect of chitosan raw material powder on the seedling height of BRRI dhan88

Seedlings treated with different levels of chitosan raw material powder significantly influenced the average seedling height of BRRI dhan88 (Fig.1). Experimental results revealed that the maximum average seedling height (17.75 cm) was obtained in C_3 (0.3 % chitosan raw material powder) treatment whereas the minimum average seedling height (13.53 cm) was obtained in C_0 (0 % chitosan raw material powder) treatment, which was statistically similar with (13.67 cm) C_5 (0.5 % chitosan raw material powder) treatment Chitosan raw material powder improved seedling growth and reduced fungi infection risk in untreated seedlings, possibly due to nutrient incorporation. The result obtained from the present study was similar to the findings of Ahmed *et al*. (2020) and they reported that seedling height was increased with the application of chitosan raw materials in the seedbed. Issak and Sultana (2017) found that *Boro* rice seedling production was improved by using the chitosan raw material powder in the seedbed.

Boonlertnirun *et al.* (2008) also reported that, chitosan is a natural biopolymer that stimulates growth and increases yield of plants as well as induces the immune system of plants.

Here, Level of chitosan raw material powder, C_0= 0%, C_1= 0.1%, C_2= 0.2%, C_3= 0.3%, C_4= 0.4% and C_5= 0.5%.

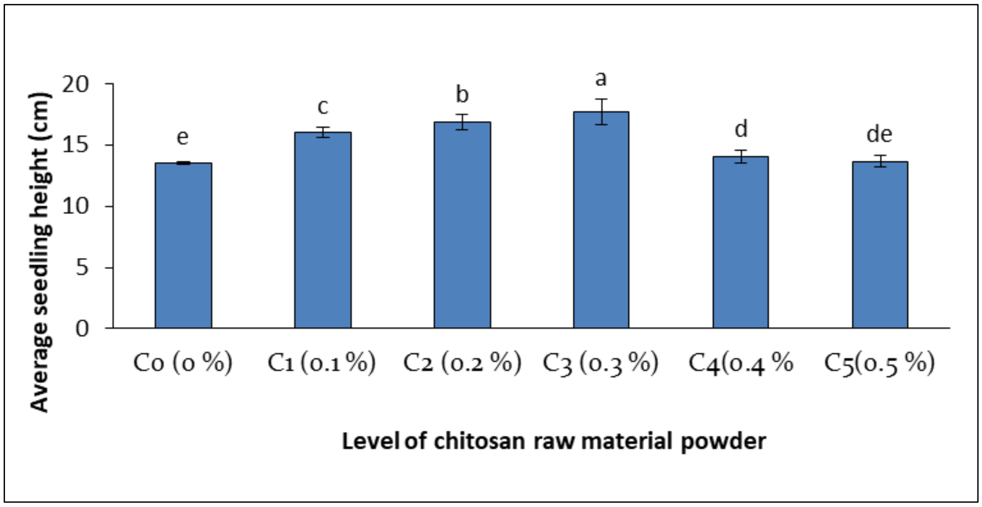


Figure 1: Effect of chitosan raw material powder level on the average seedling height of BRRI dhan88.Bars with different letters are significantly different at $p \leq 0.05$ when applying DMRT

Effect of chitosan raw material powder on fresh weight seedling^{-1} of BRRI dhan88

According to the study, adding different amounts of chitosan raw material powder had a significant effect on the fresh-weight seedling-1 of BRRI dhan88. The C_3 (0.3%) treatment yielded the maximum seedling fresh weight (531.67 mg), while the C_0 (0.5%) treatment yielded the minimum fresh weight seedling (323.58 mg). Ahmed *et al.* (2020) reported that seedling fresh weight increased with the application of chitosan-raw material powder in the seedbed. Issak and Sultana (2017) also reported that, fresh weight of BRRI dhan29 rice seedlings were influenced by the chitosan powder treatments and this might be due its supplementation of plant nutrients and growth regulators.

Here, C_0= 0%, C_1= 0.1%, C_2= 0.2%, C_3= 0.3%, C_4= 0.4% and C_5= 0.5% chitosan raw material powder

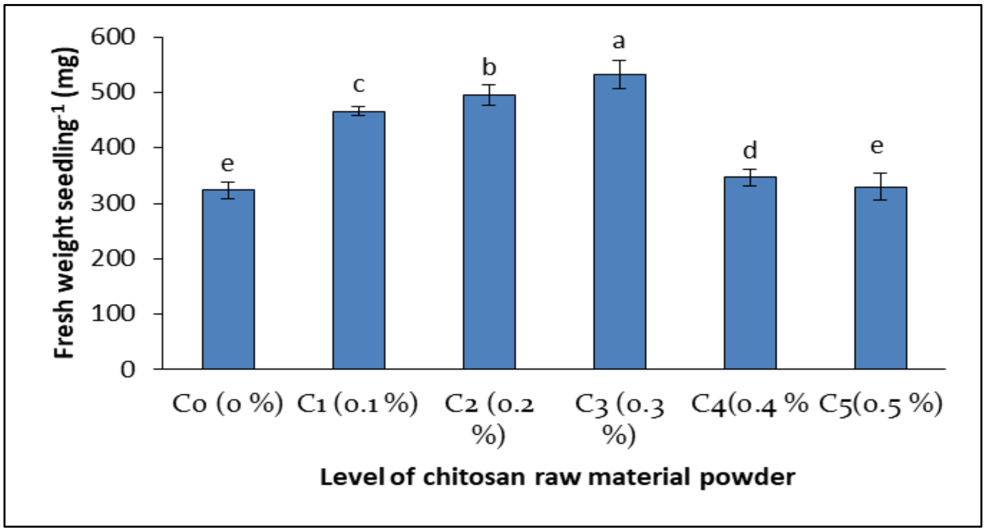
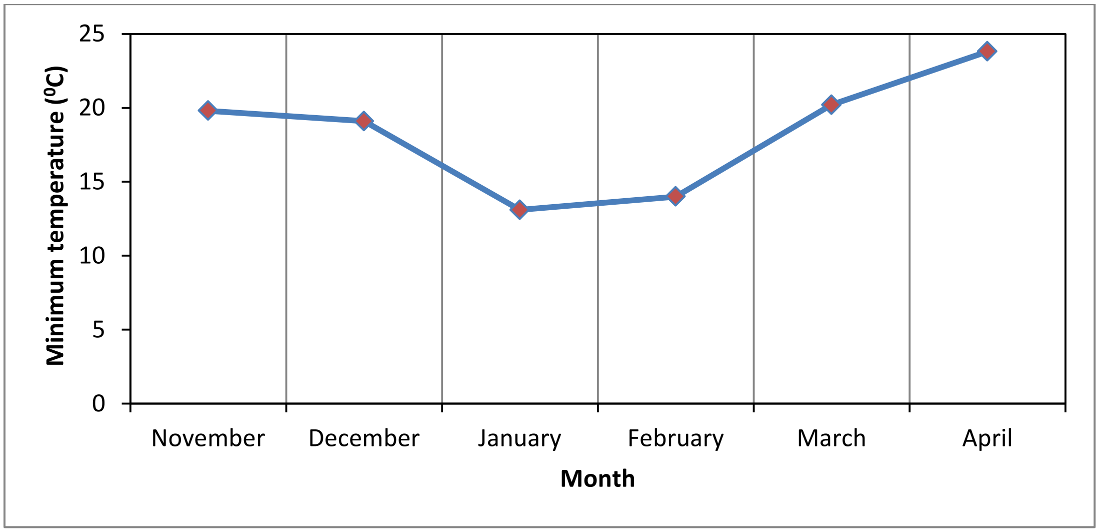


Figure 2: Effect of chitosan raw material powder level on fresh weight seedling^{-1} of BRRI dhan88.Bars with different letters are significantly different at $p \leq 0.05$ applying DMRT

Figure 3: Morphological difference of rice seedlings at transplanting stage due to the application of chitosan raw material powder



Temperature fluctuation during experimental work

Effect of chitosan raw material powder on oven dry weight seedling^{-1} of BRRI dhan88

The study found that the application of different levels of chitosan raw material powder significantly impacted the oven-dry weight seedling-1 of BRRI dhan88 (Fig. 4). The maximum dry weight seedling^{-1} was obtained in C_3 (0.3% chitosan raw material powder), while the minimum was obtained in C_0 (0.0% chitosan raw material powder). The application of chitosan raw material powder enhances plant growth and development, leading to noticeable increases in dry-weight seedling-1 compared to the control treatment. Ahmed *et al*. (2020) reported that seedling oven dry weight was increased by applying chitosan raw materials in the seedbed. Issak and Sultana (2017) also reported that Chitosan powder applications significantly influenced oven-dry weight productions of BRRI dhan29 rice seedlings, possibly due to its nutritional support, growth promotion, and potential improvement in seedbed soil properties. Boonlertnirun *et al.* (2008) found that the application of chitosan stimulates the seedling dry matter weight significantly.

Here, C_0= 0%, C_1= 0.1%, C_2= 0.2%, C_3= 0.3%, C_4= 0.4% and C_5= 0.5% chitosan raw material powder

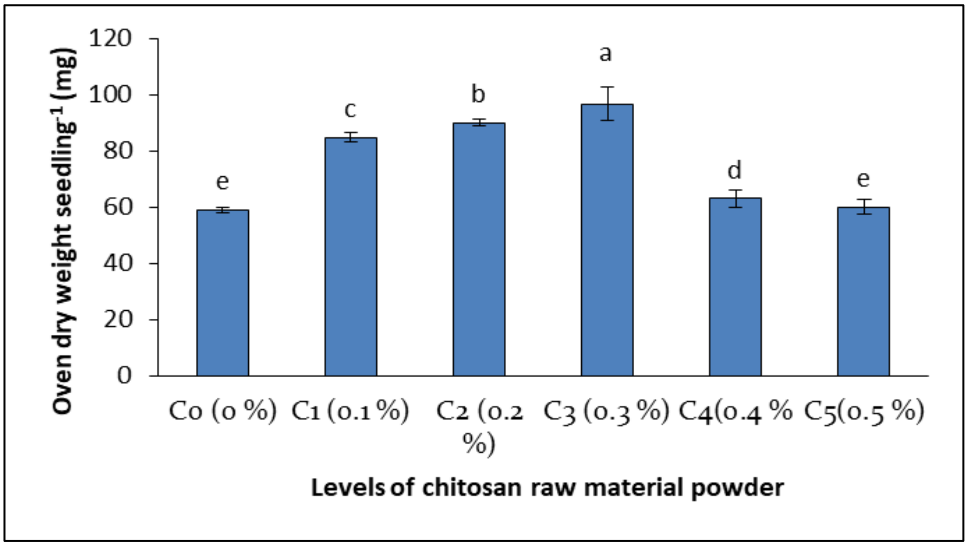


Figure 4: Effect of chitosan raw material powder level on oven dry weight seedling^{-1} of BRRI dhan88.Bars with different letters are significantly different at p ≤ 0.05 applying DMRT

Effect of chitosan raw material powder on seedling strength of BRRI dhan88

Seedling treated with different level of chitosan raw material powder significantly effect on seedling strength (mg/cm) of BRRI dhan88 (Fig. 5). Experimental result showed that, maximum seedling strength (5.44 mg/cm) was obtained in C_3 (0.3 % chitosan) treatment, whereas the minimum seedling strength (4.35 mg/cm) was obtained in C_0 (0 % chitosan raw material powder) treatment which was statistically similar with (4.38 mg/cm) C_5 (0.5 % chitosan raw material powder) treatment. Ahmed *et al.* (2020) reported that seedling strength was increased with the application of chitosan raw materials in the seedbed. Boonlertnirun *et al.* (2008) found that the application of chitosan stimulates the seedling strength significantly.

Here, C_0= 0%, C_1= 0.1%, C_2= 0.2%, C_3= 0.3%, C_4= 0.4% and C_5= 0.5% chitosan raw material powder

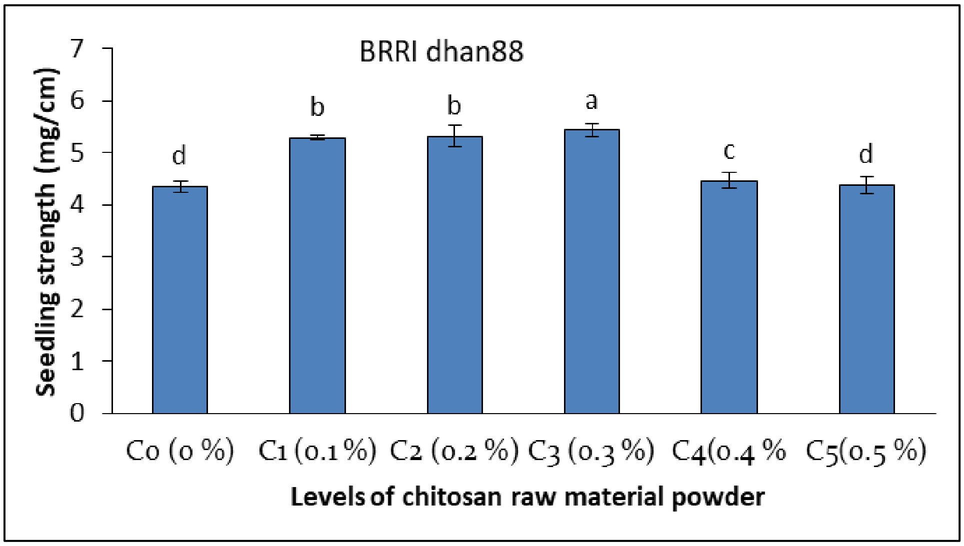


Figure 5: Effect of chitosan raw material powder level on seedling strength of BRRI dhan88. Bars with different letters are significantly different at p ≤ 0.05 when applying DMRT

Chemical Properties of Seedbed Soils after transplant

Table 3 reveals that chitosan raw material powder application levels affect seedbed soil pH and total nitrogen percentage. The C_5 treatment increased soil pH to 6.5, while the C_4 treatment recorded the highest total nitrogen at 0.12%. Chitosan raw material powder contains 7.52 % organic carbon and 12.96 % organic matter. The application of different levels of Chitosan powder affected organic carbon percentages and organic matter, with maximum levels in the C_5 treatment. However, higher amounts of organic carbon and organic matter can cause toxicity and compactness in the root zone, hindering plant growth and development

Table 3: Effect of different chitosan raw material powder treatments on pH and % total nitrogen of seedbed soil after seedling transplant

Treatments	pH and % total nitrogen				Percent organic carbon and organic matter			
	pH		% total nitrogen		Organic carbon (%)		Organic matter (%)	
	Initial	After	Initial	After	Initial	After	Initial	After
C_0	5.8	5.80 d	0.04	0.04 e	0.5	0.50 d	0.87	0.87 e
C_1	5.8	6.00 c	0.04	0.05 e	0.5	0.59 c	0.87	1.02 d
C_2	5.8	5.90 c	0.04	0.09 c	0.5	0.58 c	0.87	1.00 d
C_3	5.8	6.30 b	0.04	0.07 d	0.5	0.63 b	0.87	1.09 c
C_4	5.8	6.30 b	0.04	0.12 a	0.5	0.75 a	0.87	1.20 b
C_5	5.8	6.50 a	0.04	0.11 b	0.5	0.76 a	0.87	1.31 a
LSD$_{(0.05)}$	0	0.18	0	0.003	0	0.02	0	0.03
CV(%)	0	1.97	0	2.34	0	2.62	0	2.16

Here, Level of chitosan raw material powder, C_0= 0%, C_1= 0.1%, C_2= 0.2%, C_3= 0.3%, C_4= 0.4% and C_5= 0.5%.

Agricultural applications of chitosan can reduce environmental stress due to drought and soil deficiency, strengthen seed vigor, improve stand quality, and increase yield. By promoting root development, improving nutrient uptake, and increasing tolerance to environmental stressors, chitosan can help optimize the growth and yield potential of late-sown boro rice seedlings. Additionally, its natural antimicrobial properties can contribute to protecting the seedlings from fungal and bacterial infections, which are common during late sowing periods. However, further research and field trials are necessary to determine the optimal dosage, application methods, and environmental conditions for maximizing the benefits of chitosan in late-sown boro rice cultivation. Overall, incorporating chitosan raw powder into agricultural practices has the potential to contribute to more sustainable and resilient crop production systems.

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