TiO₂ Nanoparticles Enhancing Germination, Growth and Yield of Rice

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Authors’ contributions

This work was carried out in collaboration among all authors. Author KD undertaken the research work, carries out the bulk of the experiments, recorded all morph-physiological data in the field condition and manuscript correction. Authors AD and BD having an important contribution to experimental design, data analysis, interpretation and writing of the paper, manuscript correction, proof reading and whole correspondence during the paper submission. Author JK help in overall experimental plan and suggestion that driven the study to conclusion, manuscript correction, provided all facility and helped for preparation of this manuscript. All authors read and approved the final manuscript.

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ABSTRACT

Background: Titanium dioxide (TiO₂) NPs are used in agriculture to increase seeds germination and seedlings growth and can improve yield by approximately 30%. The present experimental investigation demonstrated the effect of nanoparticles (NPs) Anatase - TiO₂ (<100 nm size) on germination, seedling growth and yield attributing traits of rice.

Methods: Rice seeds were treated with six concentrations (0, 10, 20, 50, 80 and 100 ppm) of nanoTiO₂ and placed in Petri plates. Some amount was sown in the field following Randomized Block Design (RBD) with three replications for each treatment.

Results: Application of TiO₂ NPs significantly enhanced germination, seedling growth and yield traits for most of the concentrations as compared to the control. About 85% significant improvement

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of seed germination occurred by 20 ppm of TiO$_2$ NPs. The results of TiO$_2$ treated samples were higher in almost all the growth parameters, but the treatment was highly effective for 20 ppm followed by 50 ppm for grain yield. The response to nano-treatment on seed germination along with yield traits suggested the optimum TiO$_2$ dose limit on these characters on rice.

Keywords: Nanoparticles (NPs); TiO$_2$; rice; germination.

1. INTRODUCTION

Green revolution during 1960s, molecular breeding during 1980s and transgenic breeding highly contributed in augmentation of grain yield and amelioration of quality of rice and other crops. But such supplementary successes of crop improvement in rice in the past, their yield has now reached a plateau. To break the plateau level, novel strategies and their exploitation in complement to existing traditional and alternative advanced approaches are felt need of each hour. Nanotechnology has a great promise in transforming agriculture and food production. The development of nanodevices and nanomaterials could open up novel applications in plant biotechnology and agriculture [1]. Nanoscale devices with novel properties make the agricultural systems “smart” [2,3] and nanoparticles (NPs) generally possess dramatically different physicochemical properties compared to fine particles (FPs) of the same composition. The smaller size of NPs ensures that a large portion of atoms will be on the particle surface. Titanium dioxide (TiO$_2$) NPs are used in agriculture to increase seeds germination and seedlings growth [4] and can improve yield by approximately 30% [5]. Nano TiO$_2$ increases photosynthesis and plant growth of spinach and enhances absorption and transmission of the sun’s energy to electron energy and activates chemical energy [6]. Seeds treated with a mixture of Nano SiO$_2$ and Nano TiO$_2$ exhibited more germination and increased nitrate reductase activity in soybean [7]. It also revealed that application of nano-anatase TiO$_2$ in spinach the activity of Rubisco carboxylase was 2.67 times higher than control [8]. In this background, the aim of our approach was the application of TiO$_2$ nanoparticles by conducting experiment in the laboratory and field conditions to understanding the beneficiary effect of NPs on some physiological, morphological and yield traits of the treated rice seeds.

2. MATERIALS AND METHODS

2.1 The Experiment Sites

The experimental site was carried out at the laboratory and Instructional Farm, Bidhan Chandra Krishi Viswavidyalaya, West Bengal, India.

2.2 The Chemical and Biological Material Used

The TiO$_2$ (Anatase) nano particles (NPs) were purchased from Sigma-Aldrich Company, St.Louis, MO, USA with a purity of 99.5%, particle size of (<100 nm). A local rice variety (Oryza sativa cv. CN 1794-2) used for this experiment was procured from Regional Research Station, Chakkdah, BCKV, West Bengal. Prior to starting the experiments, rice seeds were stored in dry conditions in the dark to avoid any potential loss of their viability.

2.3 The Experimental Variants Prepared

To test the effect TiO$_2$ NPs on seed germination, the experiment was performed under laboratory conditions. Healthy seeds were selected and surface sterilized with 0.1% Mercuric Chloride solution for 1 min then vigorously rinsed with sterilized distilled water (DW) before transferring into Petri plates having a double layer of filter paper for recording germination percentage and seedling growth after 7 days. The NPs were suspended directly in distilled water and dispersed by ultrasonic vibration (100W, 40 kHz) for 30 min. Small magnetic bars were placed in the suspension for stirring before use to avoid aggregation of the particles. Six different concentration of nano TiO$_2$ (0, 10, 20, 50, 80 and 100 ppm) were prepared for seed treatment. Then, uniform and healthy seeds were treated for 48 hrs. Treated seed of the six variants were sown in Petri plates and in the field following Randomised Block Design (RBD), with three replications. Recommended manure and fertilizer were applied during field preparation.

2.4 The Studied Parameters

The seedlings parameters germination percentage, root length (cm) shoot length (cm), seedling weight (mg) and vigour index were estimated in laboratory after seven days of observation. For field experiments all the morphological and yield attributing traits were studied during maturity stage of rice.
2.5 Statistical Analysis

The results are presented as mean ± SE (standard error of the mean) for both the seedling and yield attributing traits. Each experimental value was compared to its corresponding control. Statistical significance was accepted when the probability of the result assuming the null hypothesis (p) is less than 0.05 (level of probability). The obtained data were analyzed using the SPSS software.

3. RESULTS AND DISCUSSION

The obtained results demonstrated a significantly higher germination percentage over control for all the concentration of TiO$_2$ nanoparticles. All treatments had shown maximum germination and it could be suggested that nano TiO$_2$ had not adversely affect rice seed germination. With respect to control, plants exposed with TiO$_2$ NPs showed significant improvements of germination percentage at 20 ppm (98.33%) followed by 50 ppm (98.10%) in CN-1794-2 cv. (Fig. 1). Higher concentration of TiO$_2$ treatments showed highly significant positive influence in all the seedling characters but for root length low to higher concentration was found effective, though considering the influence over the varieties higher concentration was found effective. Enhancement of root length was maximum by 50 ppm and shoot length by 80 ppm (Fig. 2). Nano TiO$_2$ showed no adverse effect on seed germination and root growth at early seedling stage [4]. All the treatments with TiO$_2$ NPs increased total seedling weight and vigour index as compared to control (Fig. 3). Lin et al. [9] observed improved water absorbing capacity, germination power and germination value, biomass, total root length from nanomaterial treated seeds. Goo et al. [8], Hong et al. [10], Zheng et al. [11] found significant positive effect on growth following nano sized TiO$_2$ treatments. Seed germination could be considered as an index of plant growth development and yield and as a beginning of the physiological processes. The effect of nanoparticles to seed germination could be related to water imbibitions that directed enhanced seed germination. The increase in seed germination along with seedling growth in specific concentrations of different nanoparticles suggested optimum dose limit on growth of seedlings. It was also found absence of any adverse effect on seed germination due to treatments with nano particles in rice [12,13]. In spinach [14] and wheat [15] revealed that accelerated germination in proper concentration of TiO$_2$ treatment and significant effect of nanosized TiO$_2$ on spinach germination is probably attributed to the small particle size, which allows its penetration into the seed during the treatment period, exerting its enhancing functions during growth.

All of the yield and its attributing characters studied, exhibited significant variation (P-values ranging from <0.001 to 0.0405) among the six TiO$_2$ concentrations (Table 1). Seed treatment with TiO$_2$ resulted in significant increase in

![Fig. 1. Effect of different concentration of TiO$_2$ NPs on germination %](image)

Bars mean the mean values ± SE
Fig. 2. Effect of different concentration of TiO$_2$ NPs on root and shoot length (cm)

Bars mean the mean values ± SE

Table 1. Effect of TiO$_2$ NPs (nanoparticles) treatment on growth and yield attributing traits of rice

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Days to 50% flowering</th>
<th>Plant height at maturity (cm)</th>
<th>Flag leaf length (cm)</th>
<th>Flag leaf breadth (cm)</th>
<th>No. of primary branches panicle$^{-1}$</th>
<th>No. of secondary branches panicle$^{-1}$</th>
<th>No. of tillers plant$^{-1}$</th>
<th>Panicle length (cm)</th>
<th>Grain yield plant$^{-1}$ (gm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>90.57$^{bc}$</td>
<td>127.77$^{b}$</td>
<td>37.77$^{a}$</td>
<td>1.070$^{a}$</td>
<td>9.82$^{b}$</td>
<td>28.67$^{b}$</td>
<td>10.85$^{a}$</td>
<td>26.69$^{a}$</td>
<td>21.51$^{b}$</td>
</tr>
<tr>
<td>10 ppm NPs</td>
<td>91.48$^{b}$</td>
<td>133.61$^{a}$</td>
<td>42.08$^{b}$</td>
<td>1.138$^{c}$</td>
<td>11.20$^{a}$</td>
<td>34.20$^{b}$</td>
<td>12.73$^{c}$</td>
<td>27.48$^{b}$</td>
<td>24.30$^{a}$</td>
</tr>
<tr>
<td>20 ppm NPs</td>
<td>85.13$^{a}$</td>
<td>134.16$^{b}$</td>
<td>40.13$^{a}$</td>
<td>1.182$^{c}$</td>
<td>9.73$^{d}$</td>
<td>29.33$^{d}$</td>
<td>14.40$^{a}$</td>
<td>29.83$^{a}$</td>
<td>25.80$^{a}$</td>
</tr>
<tr>
<td>50 ppm NPs</td>
<td>87.00$^{d}$</td>
<td>135.60$^{a}$</td>
<td>41.33$^{b}$</td>
<td>1.143$^{b}$</td>
<td>10.60$^{b}$</td>
<td>31.48$^{b}$</td>
<td>13.48$^{b}$</td>
<td>30.64$^{a}$</td>
<td>24.11$^{b}$</td>
</tr>
<tr>
<td>80 ppm NPs</td>
<td>89.60$^{c}$</td>
<td>133.37$^{a}$</td>
<td>44.11$^{b}$</td>
<td>1.170$^{b}$</td>
<td>10.37$^{c}$</td>
<td>32.89$^{b}$</td>
<td>12.13$^{c}$</td>
<td>28.53$^{b}$</td>
<td>24.43$^{b}$</td>
</tr>
<tr>
<td>100 ppm NPs</td>
<td>93.60$^{a}$</td>
<td>135.60$^{a}$</td>
<td>45.24$^{a}$</td>
<td>1.211$^{a}$</td>
<td>10.20$^{c}$</td>
<td>29.20$^{c}$</td>
<td>12.73$^{c}$</td>
<td>27.56$^{b}$</td>
<td>24.14$^{b}$</td>
</tr>
</tbody>
</table>

Means followed by a different letter within a row are significantly different at P<0.05 according to the Duncan’s multiple range test
yielding traits for all the five concentrations. It was revealed that nano TiO$_2$ has a linear effect on characters like days to first flowering and 50\% flowering at certain range, but at higher dose has a negative effect. Best response for 50\% flowering was found at 20 ppm showing earliness 6.19\% improvement over the control, respectively. An opposite result has been occurred delay in flowering and reduced seed set in plants incubated both with fullerene C70 suspended in natural organic matter solution (C70-NOM) and MWCNTs-NOM (Multwalled carbon nanotube) at a concentration of 400 mg/L in rice [14]. Plant growth, in terms of plant height was significantly increased with higher concentration of nano TiO$_2$ compared to control. The result showed that 80 ppm concentration recorded highest plant growth (135.60 cm) with 2.01\% increase followed by 100 ppm (136.63 cm) with 1.3\% increase over the control (Table 1). Flag leaf area was significantly higher in 100 ppm (54.07 cm) resulting 20.89 \% increase followed by in 80 ppm (50.45 cm) showing 12.79\% increase over the control. The result also suggested that significant positive effect on plant growth following nano sized TiO$_2$ treatments in different crops [8,11,15,16]. Yield attributing trait like number of tiller plant$^{-1}$ was significantly increased at 20 ppm with improvement of 28.57\% followed by 50 ppm. NanoTiO$_2$ treatment has a significant effect on panicle length as at 50 ppm with 3.2\% increase over the control. But at higher dose and at extreme low dose it was found a decrease of the panicle length, suggesting that an optimum concentration of nano treatment has a positive effect over the control. The primary branches panicle$^{-1}$ were found to improve with almost all the concentration except at 20 ppm, but the treatment has adverse effect on secondary branches panicle$^{-1}$. Secondary branches panicle$^{-1}$ was reduced by all the treatment showing (10 – 31\%) decrease in number. Consequently at 20 ppm the yield plant$^{-1}$ was highest resulting 25.8 g plant$^{-1}$ with 19.22\% increase as compare to control. The highly increase in yield may be attributed by increase improve in number of tillers plant$^{-1}$ and higher in primary branch.

4. CONCLUSION

The experiment suggested that the nano TiO$_2$ treatment has obvious effects on the improvement of germination, plant growth and development in rice. The effect of different concentrations of TiO$_2$ nanoparticles on the growth and yield attributing traits was maximum at 20 ppm followed by 50 ppm. The effective growth at optimum concentrations and inhibited growth beyond these may be attributed to the accumulation and uptake of nano-TiO$_2$ particles by the roots. The increased grain yield is associated with improvement of the tillers number, a greater number of primary branches and longer panicles.

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**COMPETING INTERESTS**

Authors have declared that no competing interests exist.

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