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ZnO nanoparticles affect differently the morphological and physiological responses of Riceberry plants (*Oryza sativa* L.)

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Abstract

Excessive concentrations of nanoparticles cause toxicities and harmful effects on plants and soil microorganisms. Plant species are able to tolerate to different concentrations and types of nanoparticles. The appropriation concentrations of nanoparticles could play a significant role in plant growth promotion. Therefore, this study focused on the effect of zinc dioxide (ZnO) nanoparticles on the growth rate (fresh weight, dry weight and plant height) and the contents of photosynthetic pigments (chlorophyll A, chlorophyll B and carotenoids concentrations) in Riceberry cultivar. Rice plants were treated with different concentrations of ZnO nanoparticles (0, 200, 400 and 800 mg L⁻¹) every 7 daysinterval. Rice plants were collected at 7 and 8 weeks after planting for the evaluation of plant growth parameters and photosynthetic pigment contents. The results showed that the different concentrations of ZnO nanoparticles were affected on photosynthetic pigment contents and the growth rate of Riceberry plants. The addition of ZnO nanoparticles at 200 mg L⁻¹ trended to increase the morphological and physiological characterizations when compared with other treatments. The application of ZnO nanoparticles may be applied to other rice cultivars in increasing or decreasing of seed yields, including the other plant species in the future.

Keywords: Growth rate; Photosynthetic pigments; Riceberry; ZnO nanoparticles

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1. Introduction

Nanoparticles have been shown to have higher and unique toxicity than their corresponding bulk materials [1]. Nanoparticles were found to positively or negatively affect in morphological and physiological responses. The toxicity rate of nanoparticles depends on different composition, size, concentration, and plant species [2, 3]. Fe₃O₄ and TiO₂, nanoparticles were evaluated in cucumber plants and presented negatively effect on seed germination rate, root elongation, and germination index [4]. On the other hand, Al₂O₃ nanoparticles and carbon nanotubes had significant positive effects on seed germination and growth performances of *Arabidopsis thaliana* and tomato plants, respectively [5 – 7]. Zinc oxide (ZnO) is one of metal oxide nanoparticles that have been used in agricultural research. Only a few studies have focused on their effects on plant growth and metabolism. Some studies reported that ZnO nanoparticles could be affected on plant developments and seed germinations [8, 9]. In case of rice (*Oryza sativa* L.), the previous research reported that application of ZnO nanoparticles stunted root length and reduced number of roots in rice seedlings [10]. Moreover, application of excessive concentrations of ZnO nanoparticles could be reduced the accumulation of photosynthetic pigment contents, but it contributed an activity of antioxidant enzymes in rice plants [11].

The photosynthetic pigment concentrations usually are a good indicator of plant nutrient stress, photosynthesis and growing periods. The contents of chlorophyll in the plant leave indicate the growth status and productivity of the crops. In the previous researches, ZnO nanoparticles were involved in plant toxicity and found to decline photosynthetic pigment contents. The high levels of peroxidase activities and photosynthetic pigments (chlorophyll A, chlorophyll B and carotenoids) in the rice plants may be correlated to their improved tolerance to metal nanoparticles [12, 13].

Previous studies evidenced in the both positive and negative effects of ZnO nanoparticles on plant cells. Part of the scientific literature reported their significant improvement of germinations, shoot and root growth, chlorophyll contents, transpiration and water use efficiency [14, 15]. Therefore, the effects of metal nanoparticles on the inhibition of plant growth varied greatly among types/concentrations of nanoparticles and plant species [16, 17].

In the present experiment, an attempt was made to unravel the ZnO nanoparticles induced activation of defense mechanism in rice cultivar. Therefore, the objective of this study focused on the effect of ZnO nanoparticles on plant growth performances (shoot length, root length and biomass) and photosynthetic pigment contents (chlorophyll A, chlorophyll B and carotenoids) in rice plants (Oryza sativa L. cv. Riceberry). The samples were treated with ZnO nanoparticles (0 - 800 mg L⁻¹) and endeavored in dissecting ZnO nanoparticle stress in rice plants.

2. Materials and methods

Rice plants (*Oryza sativa* L. cv. Riceberry) were used as plant materials for the present investigation. Seeds of Riceberry cultivar were soaked in tap water for 2 days at room temperature. This experiment was conducted in College of Nanotechnology, King Mongkut's Institute of Technology Ladkrabang, Thailand under the natural conditions during August-December 2016. The germinated seeds were transferred into pots containing a soil (pH 7-8) and grown in a greenhouse. Pots were maintained in a greenhouse under natural lighting and average 34 °C/25 °C \pm 3 °C day/night temperatures and the relative humidity between 50% and 65%. Rice plants were irrigated daily with tap water. Four-week-old rice plants were exposed to tap water supplemented with different concentrations of ZnO nanoparticles (0, 200, 400 and 800 mg L⁻¹) every 7 days. Rice plants were harvested at 7 and 8 weeks after planting and then stored at -80 °C until further analysis.

To evaluate the adaptation of rice plants in different concentrations of ZnO nanoparticels, the samples were measured shoot length, root length, fresh weight and dry weight for plant growth determinations. Effects of different concentrations of ZnO nanoparticles (0, 200, 400 and 800 mg L^{-1}) on physiological response were investigated in photosynthetic pigment contents that presented in the chlorophyll A (ChlA), chlorophyll B (ChlB) and total carotenoids (Car) concentrations. ChlA, ChlB and Car concentrations (mg g^{-1} FW) were measured according to the method of Lichtenthaler [18] and Shabala et al. [19]. The results were shown the mean values \pm standard deviation (S.D.) of five independent experiments (n=5) and arranged in a Completely Randomized Design (CRD).

3. Results and Discussion

Effect of ZnO nanoparticles on the plant growth performances of Riceberry cultivar

Rice plants which transplanted into a soil and irrigated with tap water supplemented with different concentrations of ZnO nanoparticles (0, 200, 400 and 800 mg L^{-1}) were used to determine the plant growth performances by measuring shoot length, root length, fresh weight and dry weight after 7 weeks and 8 weeks of transplanting.

ZnO nanoparticles caused the increase and decrease in the plant growth performances (shoot length, root length, fresh weight and dry weight) of rice plants cv. Riceberry, as shown in Fig. 1 and 2. Shoot and root lengths were slight increases in rice plants which were exposed to 400 mg L^{-1} (3.85 – 16.22% and

19.61-26.53%, respectively) and 800 mg L^{-1} (1.54-15.32% and 21.57-22.45%, respectively) of ZnO nanoparticles. Whereas shoot and root lengths showed a dramatic increases in rice plants which were exposed to 200 mg L^{-1} (20.77-23.42% and 30.61-39.22%, respectively) of ZnO nanoparticles when compared to the control treatment (0 mg L^{-1} ZnO nanoparticles) (Fig. 1).

The fresh weight and dry weight of rice plants were noted to increase among all treatments under ZnO nanoparticles conditions (Fig. 2). The fresh weight and dry weight observed for rice plants dramatically increased from 32.51 to 88.89% and 49.56 to 73.62%, respectively when exposed to 200 mg L^{-1} ZnO nanoparticles. Whereas the rice plants demonstrated a slight elevated in 400 mg L^{-1} (5.28 – 20.16% and 15.95 – 43.36%, respectively) and 800 mg L^{-1} (8.89 – 9.47% and 20.25 – 38.05%, respectively) of ZnO nanoparticles treatments as well as the results of shoot and root length performances.

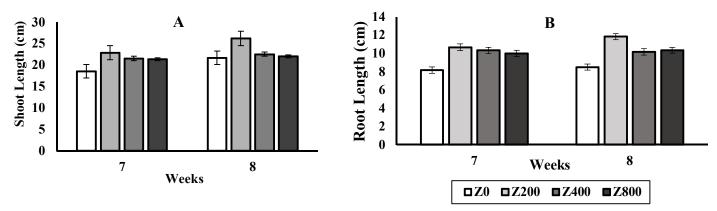


Fig. 1 The effects of different concentrations of ZnO nanoparticles [0 (Z0), 200 (Z200), 400 (Z400) and $800 (Z800) \text{ mg L}^{-1}$] on (A) shoot length and (B) root length of rice plants cv. Riceberry. Data are expressed as the mean values \pm standard deviation (S.D.) (n = 5).

Effect of ZnO nanoparticles on the photosynthetic pigments of Riceberry cultivar

Increments of ZnO nanoparticles concentrations were presented a reduction of augmentation rate in physiological parameters (ChlA, ChlB and Car concentrations). In this study, 200 mg L^{-1} ZnO nanoparticles treatment obviously induced a high increase in ChlA, ChlB and Car concentrations (61.20 – 88.23%, 104.06 – 116.80% and 66.45 – 79.19%, respectively) relative to the level in the control treatment (0 mg L^{-1} ZnO nanoparticles) (Fig. 3). A slight increment in ChlA, ChlB and Car concentrations was observed in 400 mg L^{-1} (0.59-27.47%, 37.58 – 68.19% and 21.24 – 39.38%, respectively) of ZnO nanoparticles. Moreover, ChlB and Car concentrations were slightly increased in 800 mg L^{-1} (18.33 – 39.15% and 17.10 – 19.12%, respectively) ZnO nanoparticles treatment of rice plants. Whereas ChlA concentrations of rice plants exposed to 800 mg L^{-1} (1.49 – 8.26%) ZnO nanoparticles were slightly decreased when compared to the control treatment (Fig. 3).

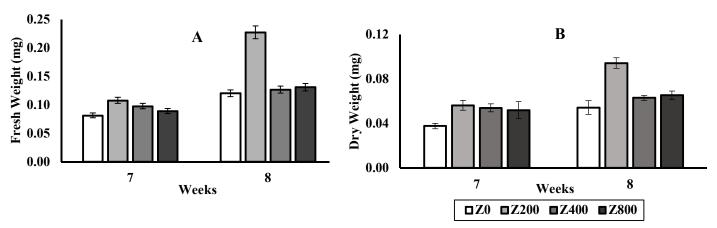


Fig. 2 The effects of different concentrations of ZnO nanoparticles [0 (Z0), 200 (Z200), 400 (Z400) and 800 (Z800) mg L⁻¹] on (A) fresh weight and (B) dry weight of rice plants cv. Riceberry. Data are expressed as the mean values ± standard deviation (S.D.) (n = 5).

The enhancements of morphological parameters (the plant growth performances) were found in rice plants exposed to 200 mg L⁻¹ ZnO nanoparticles which were correlated with physiological parameters (the photosynthetic pigments). The results showed that that 200 mg L⁻¹ ZnO nanoparticles treatment gave higher plant growth performances and the photosynthetic pigment contents than other treatments (0, 400 and 800 mg L⁻¹ ZnO nanoparticles). The previous researches demonstrated that the several of plant species differently response to various concentrations and exposure time of ZnO nanoparticles [20, 21]. The results of Garcia-Gomez et al. [22] and Kim et al. [23] studies found that ZnO nanoparticles significantly reduced and induced the plant growth and plant biomass accumulation; however the toxic of ZnO nanoparticles were depended on the concentrations and/or period of ZnO nanoparticles exposure. The ZnO nanoparticles highly dissolve in soils and significantly increase in solubility in mediums when compared with the bulk ZnO due to it got high specific surface area [24, 25].

The photosynthetic pigment contents is considered as the important indicators for detection of toxicity characterization in plant cells that induced by abiotic stress such as heavy metal or oxidative condition [26, 27]. The increased growth rate and biomass in plants might be due to enhanced accumulation of photosynthetic pigment contents during the photosynthetic process. Besides, the enhanced accumulation of photosynthetic pigment contents might be affect on the amount of plant productivity [28-30].

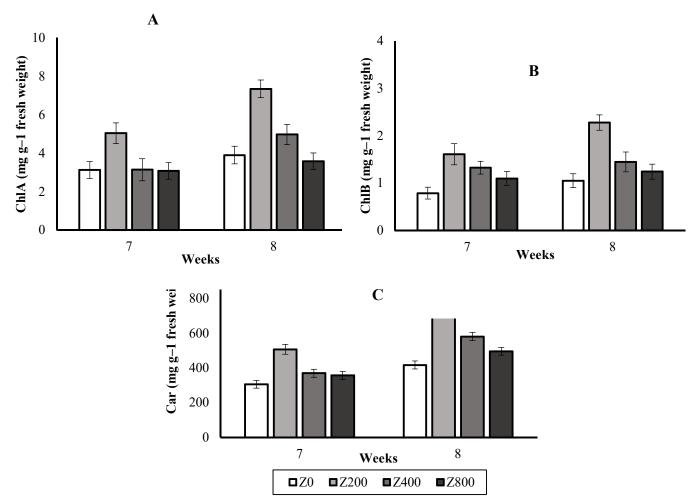


Fig. 3 The effects of different concentrations of ZnO nanoparticles [0 (Z0), 200 (Z200), 400 (Z400) and 800 (Z800) mg L^{-1}] on (A) chlorophyll A (ChlA), (B) chlorophyll B (ChlB) and (C) carotenoid (Car) concentrations of rice plants cv. Riceberry. Data are expressed as the mean values \pm standard deviation (S.D.) (n = 5).

4. Conclusion

This study focused on the effect of different concentrations of ZnO nanoparticles on the plant growth performances (shoot length, root length, fresh weight and dry weight) and the photosynthetic pigment contents (ChlA, ChlB and Car concentrations) in rice plants cv. Riceberry. The results could be concluded that the application of ZnO nanoparticles was significant difference for the plant growth and photosynthetic pigment contents in rice plants. Enhanced the high plant growth and the photosynthetic pigment contents were found in rice plants cv. Riceberry exposed to ZnO nanoparticles treatments, especially at low concentrations of ZnO nanoparticles (200 mg L⁻¹).

5. Suggestions

In the future, the effect of different concentrations of ZnO nanoparticles on the yield of rice should be studied. Moreover, the relationship between ZnO nanoparticles and the different cultivars of Thai rice are interesting to study further.

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