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Growth Response of Wheat to Titania Nanoparticles Application

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Abstract

Advancement in nanotechnology and its impacts have raised concerns about the application of engineered nanomaterials in agriculture and environment, especially on plants. In this work, the effects of TiO_2 nanoparticles (TiO_2NPs) were investigated on growth of wheat plants (Triticum aestivum). Main focus was to evaluate the effects of TiO_2 NPs on plant's morphological parameters like root, shoot length and biomass. The seeds of wheat were sown in soil containing different concentrations of TiO_2 NPs i.e. 0, 20, 40, 60, 80, 100 mg/kg. There were four replicates for each treatment. A random block design was used to carry out the experiments at IESE, NUST Islamabad, Pakistan. The SEM-EDX analysis was performed to observe the uptake of TiO_2 NPs by the wheat plants. The results showed that root, shoot length and biomass were significantly affected by TiO_2 NPs treatments. An increase in the plant's root and shoot lengths and, biomass was observed up to 60 mg/kg of TiO_2 NPs. Higher concentrations of TiO_2 NPs not only affected the root and shoot lengths but also reduced its biomass. The results of this study suggest that further investigations should be made to determine the possible consequences and impacts of applying NPs on other agricultural crops as well.

Keywords: Wheat, Titania, Growth response, Biomass, uptake, Toxicity

Introduction

Nanoparticles (NPs) have average size of less than 100 nm and have unique properties that depend on their phase, distribution, size and morphology [1]. These NPs have gained much attention in a number of consumer products, cosmetics, transportation, pharmaceutics, energy and agriculture. The environmental impacts of these materials are rarely studied and reported in literature.

Titanium dioxide nanoparticles (TiO_2NPs) are extensively utilized in composition of numerous commercial products such as paints, cosmetics, sunscreens, surface coatings and pigments [2], and also for environmental remediation e.g. soil, water and air [3,4]. According to previous computer modeling assessments, this large production and utilization may lead to their release in the environment and about 136 mg TiO_2NPs would be present in per kg of sewage sludge [5]. This sludge if applied to agricultural lands, may affect the soil and the plants health in the long run. Crops grown on these soils are more likely to accumulate TiO_2 NPs in their roots and can translocate them to their shoot. It has been reported that NPs are taken up by plant roots and sometimes translocated to their seeds, leaves and fruits [6,7]. This accumulation, however, depends on the properties of NPs (shape, size, chemical composition, agglomeration) and plant species [8]. Some studies have reported the positive effects [9 -11] while others have found the negative effects [12 - 14] of TiO_2NPs on plants. For these reasons, there is a need to further study the possible effects of these NPs on plants, especially grain crops because of their greatest concern for the human health.

In this context, the objectives of present study were: i) to assess the effects of TiO_2 NPs on root and shoot lengths, total fresh and dry biomass of plants, ii) to observe the uptake of Ti in roots and leaves by scanning electron microscope (SEM) and mapped by energy dispersive x-ray spectroscopy (EDX). Wheat (Triticum aestivum) was used as test plant species because of its widesprea

Materials and Methods

Characteristics of TiO_2NPs and Experimental Soil

TiO_2NPs were prepared using Liquid Impregnation method [15]. The surface morphology of TiO_2 NPs was explored by SEM (model: JSM-6490A, JEOL Japan). The image of the pure titania showed that particles were spherical in shape and distributed in the range of 11.93-18.67 nm (Fig. 1).

The soil was collected from the vicinity of the institute and left over 3 days for air drying. Afterwards, soil was pulverized by Ball Mill and sieved to remove pebbles, roots and vegetation. The processed soil (≤ 2 mm particle size) was used in experimentation. Soil texture was determined using saturation percentage method [16] and characterized as sandy loam having pH 7.6. d production and importance as food crop in the world.

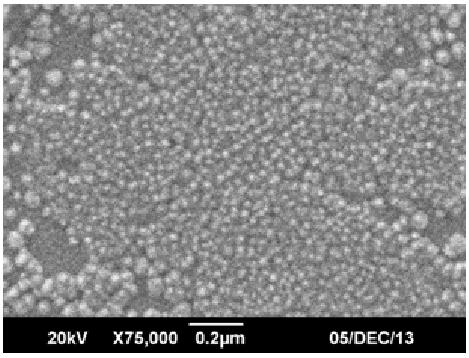


Fig. 1: XRD pattern of synthesized TiO_2 NPs

Seed Germination and Exposure

Five concentrations of TiO_2NPs were prepared, 20, 40, 60, 80, 100 mg/kg of soil. In every experiment, control (soil without NPs) group was also set for comparison with the treated ones. Different concentrations of TiO_2NPs were mixed manually in 300 g soil for each replicate and then pots were filled with TiO_2NPs mixed soil. Distilled water was added in soil and then pots were kept for 24h for NPs stabilization.

Healthy seeds of variety 'Galaxy 2013' of wheat were obtained from the Ayub Agricultural Research Institute (AARI), Faisalabad, Pakistan. The seeds were rinsed and immersed for three hours in distilled water. After that, three seeds were sown in each pot. All the experiments were repeated three times to be sure for the findings of this study. Each treatment had four replications.

Determination of Plant Growth

Plants were harvested after 60 days exposure time to investigate the effects of TiO_2 NPs on root and shoot lengths, total fresh and dry biomass of plant. For analysis, roots were washed with distilled water carefully to remove soil, and shoots were cut and their lengths were taken. After this, fresh biomass was measured one by one. To examine the dry biomass, these were kept in glass Petri dishes for oven drying at 80°C. The dry biomass was recorded after 48h.

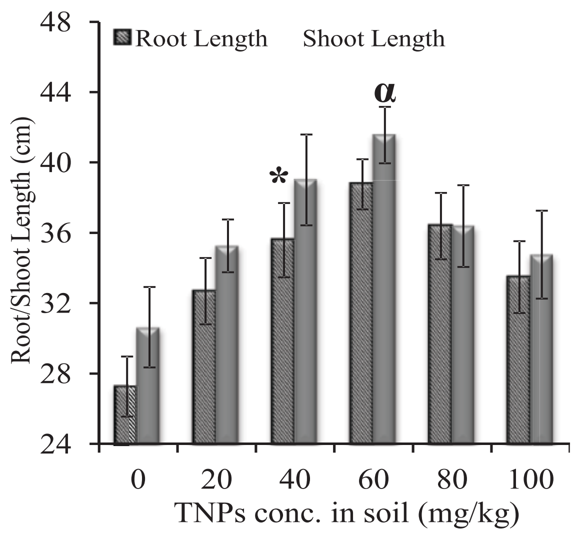


Fig. 2: TiO_2 NPs effects on root and shoot length of wheat. Vertical bars show SD and asterisk symbol (*) and α represent statistically significant difference at $p< 0.05$.

Ti uptake in Plants

The images of thin sections of dried roots and shoots of wheat were acquired independently using SEM, equipped with EDX.

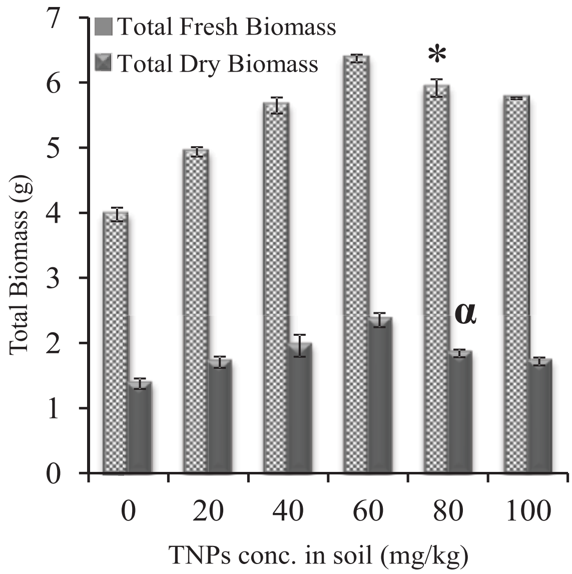
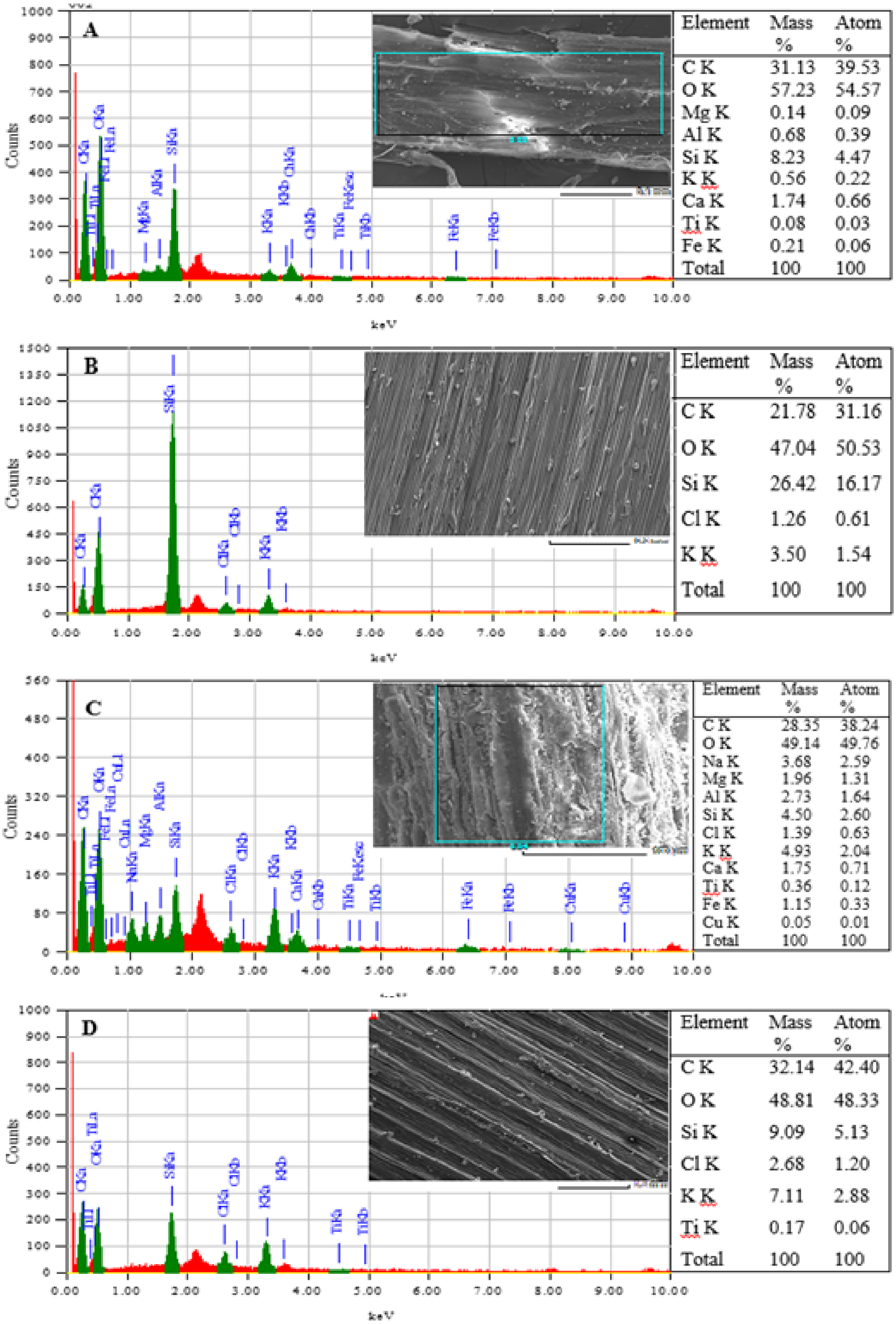


Fig. 3: Effects of TiO_2 NPs on biomass of wheat. Error bars indicate SD and asterisk symbol (*) and α show statistically significant difference at $p< 0.05$.

Results and Discussion

The effects of TiO_2 NPs on root and shoot lengths are shown in Fig. 2. The root and shoot length of wheat was increased with increasing concentration of TiO_2 NPs when the concentration was lower than 60 mg/kg, but the root and shoot length was decreased when the concentration of TiO_2 NPs was higher than 60 mg/kg. The root and shoot lengths were increased significantly ($p< 0.05$) by 48% and 40.3% respectively at 60 mg/kg in comparison with control and then followed by a gradual decrease at concentrations of 80 and 100 mg/kg TiO_2NPs. It is reported that the high surface reactivity of TiO_2NPs might enlarge root pores and in turn, water absorption and nutrients availability to plants is improved [8]. However, increasing concentration of TiO_2NPs caused aggregation of NPs that might decrease the water availability due to clogging of root pores [17]. Reports in literature indicated that TiO_2NPs had positive effects at suitable concentration and had negative effects in high concentrations [18].



A

Element	Mass %	Atom %
C K	31.13	39.53
O K	57.23	54.57
Mg K	0.14	0.09
Al K	0.68	0.39
Si K	8.23	4.47
K K	0.56	0.22
Ca K	1.74	0.66
Ti K	0.08	0.03
Fe K	0.21	0.06
Total	100	100

B

Element	Mass %	Atom %
C K	21.78	31.16
O K	47.04	50.53
Si K	26.42	16.17
Cl K	1.26	0.61
K K	3.50	1.54
Total	100	100

C

Element	Mass %	Atom %
C K	28.35	38.24
O K	49.14	49.76
Na K	3.68	2.59
Mg K	1.96	1.31
Al K	2.73	1.64
Si K	4.50	2.60
Cl K	1.39	0.63
K K	4.93	2.04
Ca K	1.75	0.71
Ti K	0.36	0.12
Fe K	1.15	0.33
Cu K	0.05	0.01
Total	100	100

D

Element	Mass %	Atom %
C K	32.14	42.40
O K	48.81	48.33
Si K	9.09	5.13
Cl K	2.68	1.20
K K	7.11	2.88
Ti K	0.17	0.06
Total	100	100

Fig. 4: Uptake of TiO2NPs in root and shoot of wheat analyzed by SEM and EDX. A & B) roots and leaves treated with no TiO2NPs, C & D) roots and leaves when treated with 100 mg/kg TiO2NPs. The peaks show Ti mass% and atom% in the sample.

The total fresh and dry biomass of wheat was increased with increase in the concentration of TiO_2NPs up to 60 mg/kg but decreased at higher treatment levels (Fig. 3). In comparison with control, plants treated with TiO_2NPs showed improvements in total fresh (60% increase) and dry biomass (72% increase) significantly ($p < 0.05$) at 60 mg/kg. Zheng et al. [19] reported that the best results of spinach growth were observed at low concentration levels (< 2500 mg/L) of TiO_2NPs as compared to high concentration levels (> 2500 mg/L).

The total fresh and dry biomass showed the same trend as root and shoot length of wheat when exposed to TiO_2NPs. It can be explained by the fact that plant is composed of roots and shoots, and their growth changes will affect the biomass. It is postulated from the results that TiO_2NPs at 60 mg/kg proved to be more effective as compared to low and high treatment levels. It is also possible that wheat is more sensitive and permits the addition of TiO_2NPs in a limited range i.e. 20-60 mg/kg in our case.

After 60 days exposure time, Ti uptake was studied by SEM and EDX on thin section of roots and leaves of wheat. The results (Fig. 4A & B) showed that the roots of control plants also contained traces of Ti, this might come from naturally occurring Ti in soils [20], whereas Fig. 4C and D displayed the images of roots and leaves of wheat when treated with 100 mg/kg of TiO_2NPs. The results demonstrated that TiO_2NPs with diameter ranging from 11.93-18.67 nm were observed in plants' roots and translocated to leaves upon root exposure. The uptake of Ti increased with increasing concentration level of TiO2NPs. According to the previous study, TiO2NPs of less than 36 nm are taken up by roots and accumulated in root parenchyma of wheat. Due to the small size, these NPs cross the Casparian band and reach vascular cylinder. It is possible that the accumulation of TiO_2 NPs in roots may alter the cell wall structure and induce production of reactive oxygen species (ROS) [8]. TiO_2NPs induced the production of oxygenated free radicals by reaction with dissolved oxygen and water when exposed to UV light and in dark also [21]. The production of oxidative species (e.g. H_2O_2) caused by NPs exposure may result into many harmful effects to plant cells including DNA damages [22]. However, this exposure-response could be species dependent as lettuce plants (Lactuca sativa) had not shown any negative impacts 100 mg/kg TiO_2NPs and a continuous increase in plant growth was observed [23].

Conclusions

Titanium dioxide NPs affect the growth of wheat but the effects are positive up to 60 mg/kg concentration levels. TiO_2NPs increased root and shoot length and total fresh and dry biomass up to 60 mg/kg but decreased at higher concentrations. Wheat accumulated and translocated Ti to leaves due to soil application of TiO_2NPs. Based upon the results, it can be concluded that TiO2NPs may have inhibitory effect and cause cell damage at concentrations higher than 60 mg/kg TiO_2 NPs. This establishes the need to further investigate the possible consequences and impacts of applying other NPs (such as ZnO NPs) on agricultural crops.

Acknowledgement

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