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Application of silver nanoparticles to improve wheat seedlings growth

A A Asanova[1], S E Yashin[1], T V Trofimova[2] and V I Polonskiy[1,2]

[1] Krasnoyarsk State Agricultural University, 90 Mira st., 660049, Krasnoyarsk, Russia
[2] Siberian Federal University, 79 Svobodniy av., 660041, Krasnoyarsk, Russia

E-mail: nastasia.asanova@gmail.com

Abstract. In the current study, the effects of silver nanoparticles (AgNPs) synthesized under the action of "green" reagent glucose, on seed germination and seedling growth of wheat were investigated. Nanoparticles had a spherical shape and an average size of about 10 nm. Wheat seeds and seedlings were cultivated in Petri dishes for 3-7 days. After this, the number of germinated seeds, shoot and root fresh mass, and root length were measured. The results showed that the concentration range of 0.001-0.5 mg/L had no effect on seed germination, while the enhanced concentration induced an inhibitory effect. It was found that AgNPs had led to an increase in shoot and root fresh mass in the range of 0.06-1 mg/L and in the range of 0.03-0.1 mg/L, respectively. Beyond the concentrations up to 3 and 5 mg/L shoot and root fresh mass were inhibited respectively. Application of tested nanoparticles in the range of 0.005-0.5 mg/L caused an increase in root length of wheat. AgNPs at the concentration over 5 mg/L caused root length inhibition. The results show that AgNPs in the range of 0.06-0.5 mg/L may hold significant applications in agriculture and could provide as an alternative source of ecofriendly fertilizer for wheat.

1. Introduction

Due to the rapid growth of agricultural products, one of the problems of the present world is the environmental damage caused by agrochemical technologies. Therefore, the use of nanotechnologies in crop production is one of the most promising research areas at the present time. Among the driving factors for yield and crop quality is the plant growth and development stimulation with an active ingredient. Such active ingredients are silver nanoparticles (AgNPs) which have prolonged action and do not require the large doses to achieve the desired biological effect.

To date, it has been mainly reported negative effects of AgNP on plants [1,2,3] including wheat [4 5,6,7]. There are only a few data dedicated to the positive effect of AgNPs on plants. Some studies have indicated that AgNPs can improve seed germination and seedling growth in Boswellia ovalifoliolata [8] and Pennisetum glaucum [9] and seedling growth in Brassica juncea [10], Panicum virgatum, Phytolacca americana [2], Phaseolus vulgaris and Zea mays [11].

It is expected that the determining dose response of plants to nanoparticles will improve our ability to use AgNPs to increase crop yields. Therefore, the purpose of this study was to investigate the AgNPs effect on seed germination and seedling growth of wheat to assess their future usage in agriculture as a plant growth promoter.

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2. Materials and methods

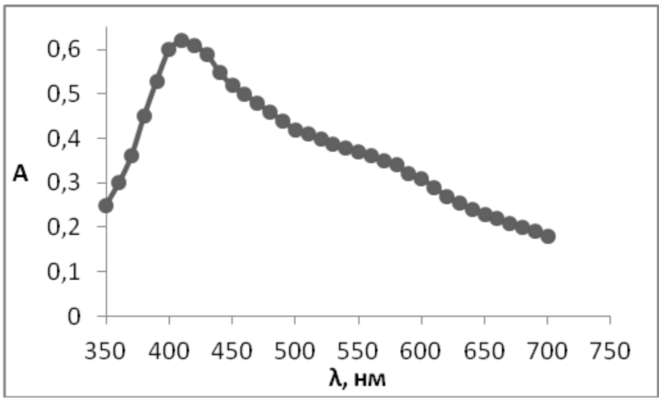
The experiments were carried out using AgNPs, which have been synthesized by reducing Ag-ion under the action of "green" reagent glucose without additional stabilizer substance. According to the method [12], to 10 ml of 0.0001 M silver nitrate solution (LenReactive) was added the same volume of reducing agent, 0.05 M glucose solution (LenReactive). The pH value was adjusted to level of 8 using ammonia solution. The obtained solutions were processed in a microwave oven for 10 minutes at a power of 700 W, which ensured uniform and rapid heating of the entire volume of reaction solution and as a result, uniform size and shape nanoparticles. The optical absorption spectrum of silver sols was recorded in the 300–700 nm range on a UV-1200 spectrophotometer (TM EKOVYU). Micrographs of samples were taken on a Hitachi 7700M electron microscope at an accelerating voltage of 30 kV.

The stimulating effect of AgNPs was determined on seed germination and seedling growth of higher plant Triticum aestivum L. Non-deformed and healthy wheat seeds with germination rate not less than 80% were selected for the experiment. Seed germination and seedling growth were carried out in Petri dishes. 25 dry seeds were placed in each dish for the experiment. Next silver sols or a glucose solution as a control were added in a volume of 5 mL to the seeds. Closed Petri dishes were thermostated at a temperature of 20-23° C for 3 days. After counting the number of germinated seeds, all samples were placed back into the thermostat so that the total incubation time is 7 days. At the end of the exposure period, the root and shoot fresh mass and the root length in the control and experimental samples were measured. When the root length measuring, the object of study was the root with the maximum length.

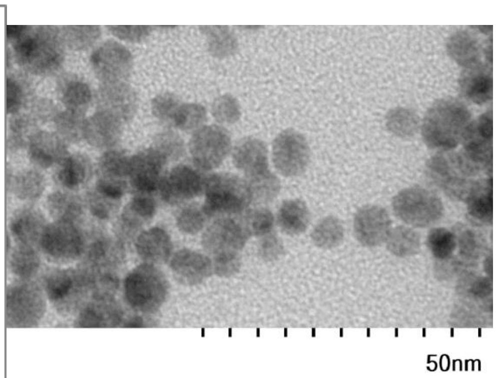
Means and standard deviations were derived from measurements on three replicates for each treatment and the related controls. The data obtained from the various treatments were statistically analysed using the t-test at a significance level of 0.5.

3. Results and discussion

During the synthesis of NPs, according to the position and intensity of the maximum surface plasmon resonance (λmax = 410 nm) in the optical spectrum (figure 1a), the stable colloidal silver particles of yellow color and spherical form was established. According to transmission electron microscopy (TEM) data, particles had uniform spherical shape with an average size of about 10 nm in diameter (figure 1b).



a)



b)

Figure 1. a - optical spectrum of AgNPs (λmax = 410 nm), b - TEM image of silver nanoparticles.

We investigated the AgNP effects in the range of 0.001 to 10 mg/L on seed germination and seedling growth of Triticum aestivum L. At the low concentration range of 0.001 to 0.5 mg/L AgNPs were shown to have no significant effects on seed germination. When the concentration of 0.5 mg/L was exceeded it was observed inhibition of seed germination. The stimulating effect of AgNPs was determined on shoot fresh mass within the range of 0.06 to 1 mg/L. Nanoparticle concentration of 0.7

mg/L was found to maximum stimulation shoot fresh mass more than two times (212%). Shoot fresh mass inhibition has been demonstrated above 3 mg/L (figure 2). AgNP dosage from 0.03 to 0.7 mg/L was found to stimulate root fresh mass of seedlings, while the concentrations above 5 mg/L induced an inhibitory effect. The maximum root fresh mass (144 %) observed in seedlings treated with 0.33 mg/L (figure 2).

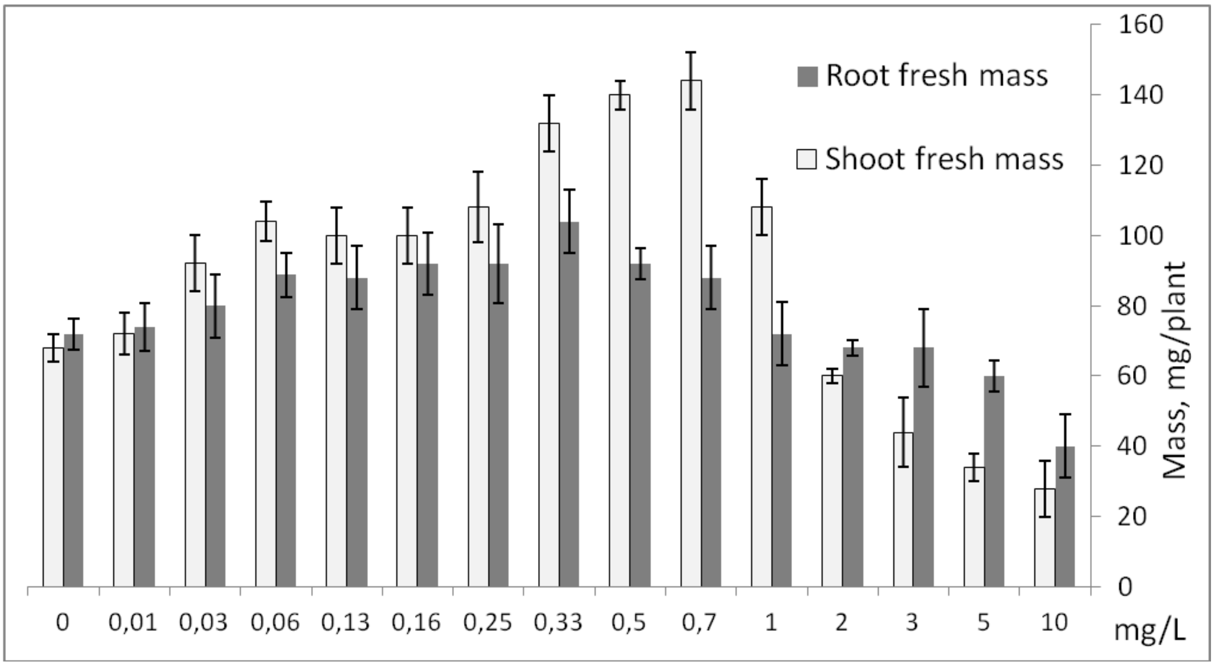


Figure 2. Effect of AgNP on shoot and root fresh mass of wheat on day 7 (Mean±SE).

The root length of seedlings was positively affected in the range of 0.005 to 0.5 mg/L. Concentration of 0.06 mg/L was found to maximum induce a 187% increase in root length of the treated seedlings (figure 3), whereas treatment with higher concentrations of 5 mg/L was found to decrease root length of seedlings.

Other authors claimed negative effects of AgNPs on wheat. Results in [4] reported that plant growth and wheat ripening were significantly inhibited when they were grown in three different concentrations (20, 200, 2000 mg/kg) of AgNPs amended soil for 4 months. Other studies demonstrate that interconversions of Ag forms outside and inside the plant, leading to a mixture of Ag species. It was observed that the root tip cells of wheat can readily internalize the AgNPs and the internalized AgNPs can cause genotoxic effect and interfere with the cells' normal function [5, 7]. It was found morphology disorder, damage to cell membranes, severe oxidative stress and in consequence intensification of production of non-enzymatic antioxidants in the concentration range between 20 and 60 ppm (20-60 mg/L) in treated seeds [6].

A number of studies dedicated to other plants reported a positive effect of AgNPs. It was observed a higher percentage of seed germination of Boswellia ovalifoliolata and it took less time when compare to control. The maximum height (10.6 cm) observed in seedlings treated with 4 mg/L [8]. It was reported in [9] that AgNPs did not adversely affect the seed living process of Pennisetum glaucum and infact they had enhanced seed germination up to 93.33 %. Fresh weight, root and shoot length, and vigor index of 7-day-old Brassica juncea seedlings were positively affected by AgNP treatment in the range of 20-400 ppm (20-60 mg/L). It induced a 326 % increase in root length and 133 % increase in vigor index of the treated seedlings. Improved photosynthetic quantum efficiency and higher chlorophyll contents were recorded in leaves of treated seedlings, as compared to the control seedlings [10].

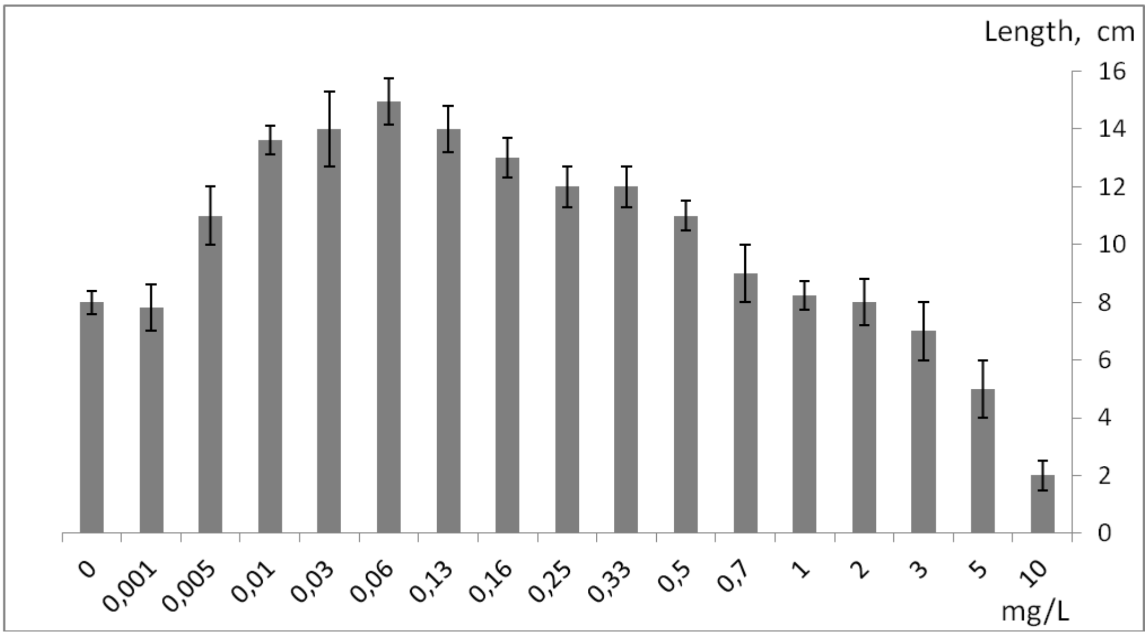


Figure 3. Effect of AgNP on root length of wheat on day 7 (Mean±SE).

Concentrations of AgNPs from 20 to 60 ppm (from 20 to 60 mg/L) had led to an increase in shoot and root lengths, leaf surface area, chlorophyll, carbohydrate and protein contents of Phaseolus vulgaris and Zea mays [11]. AgNPs treatments elicited significant positive effect on Carex lurida leaf growth at a concentration of 40 mg/L. Phytolacca americana and Panicum virgatum, responded to AgNPs treatments by growing significantly longer and more curved roots compared to the control at the same concentration of 40 mg/L [2]. The tulips treated with 100 mg/L AgNPs flowered earlier, had longer cut-flower stem, larger petals, greater stem diameter and cut-flower fresh weight [13].

The results obtained in our study and literature analysis show that the impact of AgNPs on higher plants primarily appears to depend on the AgNP dosage. Exposure to specific range of concentrations of AgNPs could improve plant growth but higher concentrations could affect negatively. In addition, other characteristics of nanoparticles such as size, reducing agent and the stabilizing substance could impact on nanoparticle toxicity. So using the roots of Allium cepa it was shown that AgNPs coated with cetyltrimethylammonium bromide had the highest toxicity compared to nanoparticles coated with citrate and polyvinylpyrrolidone [1]. In other work on higher plants it was found that the effect of nanoparticles varies greatly from both the test object and different coatings of nanoparticles [2].

4. Conclusion

AgNPs on the studied concentration range from 0.001 to 10 mg/L did not have a stimulating effect on germination energy of wheat seeds. In the range of 0.06-0.5 mg/L, the root and shoot fresh mass and the root length had a positive effect. At a concentration of 5 mg/L, inhibition of the initial growth processes was observed for all the test functions of wheat. The most sensitive parameter was the root length. The maximum increase (187 %) was recorded at a concentration of 0.06 mg/L. The maximum stimulating effect (212%) was achieved on shoot fresh mass at a concentration of 0.7 mg/L. These results revealed that AgNPs synthesized under the action of “green” reagent glucose can provide significantly enhance root and shoot fresh mass and root length. It could be used to efficient and ecofriendly strategy for growth improving of higher plants.

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