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The effect of nanoparticles of biogenic ferrihydrite on the Lemna minor L. growth

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Abstract. The article presents the data of experimental studies to identify the effect of biogenic cobalt-doped ferrihydrite nanoparticles on the growth of a model test plant Lemna minor. The test object was exposed for 7 days in a Steinberg medium with the addition of nanoparticles suspension (1.5 mg/l, 4.5 mg/l, and 15.0 mg/l). The analysis of the experimental data showed a suppressive effect of biogenic cobalt-doped ferrihydrite nanoparticles on the number of fronds and the growth rate of Lemna minor at a concentration of 15.0 mg/l. The indicators of root growth (number, length, weight) were similar to the control ones. The obtained data confirm the available data about the negative impact of elements in the form of nanomaterials at high concentrations and require further studies.

1. Introduction

In connection with the intensive use of nanomaterials and nanotechnologies, the study of the biological effect of metal-containing particles not only of technogenic [1, 3], but also of biogenic origin [2, 3, 4] presents certain interest. Iron is an essential element for organisms. This metal is spread in biological habitats. Iron is absorbed in large quantities by plants therefore plants composition contains up to 0.08 % of iron. According to physiological functions, iron is a typical trace element, found in water and soil; iron is a part of a number of compounds, including ferrihydrite [5]. Currently, it is possible to use iron preparation at the level of nanoparticles. Nanoparticles have a high adsorption capacity and high transport activity that determines a real ground of impact on plant organisms [6], using minimal doses of elements. It has been proved that in natural environment nanoparticles are produced biogenically by a number of bacteria, living in the bottom sediments of water basins [7]. Actual studies of the properties of ferrihydrite nanoparticles both in pure form [8] and in the form of nanoparticles, doped with various elements are carried out on a number of organisms [3]. The expansion of the scope of nanoparticles application determines the need to study the reaction of various standard test objects on ferrihydrite nanoparticles to identify a possible toxic effect.

2. Methods

The duckweed species Lemna minor is used as a model organism for higher water plants [9]. Duckweeds are monocotyledonous, free-floating angiosperms and belong to the Arales within the subclass of Aridae. Duckweeds are fast growing higher plants, spreading from the tropic to the



arctic zone. As primary producers they are a food source for waterfowl, fishes, small animals and serve as physical support for a variety of small invertebrates.

Duckweed can be damaged by water constituents and effluents. The subsequent inhibition of growth is calculated from the observation parameters (frond number, frond area, chlorophyll, dry weight) by a number of defined calculation methods [10].

Laboratory studies were carried out using climatic chamber with the regulation of external factors (temperature 27 °C, lighting 3-4 Klux, humidity 80 %). *Lemna minor* L. was grown on 100 % of the nutritional medium of Steinberg [10].

Three-frond organisms placed in 50 ml vials were used in the experiments. Biogenic cobalt-doped ferrihydrite nanoparticles in the form of suspension were added to the test medium with 2 % of Steinberg nutrient medium to concentrations of 1.5 mg/l, 4.5 mg/l, 15.0 mg/l.

Nanoparticles were used in the form of stable aqueous sol, obtained by the technology described in the paper [7]. The nanoparticles are iron hydroxide (ferrihydrite, $5\text{Fe}_2\text{O}_3 \cdot 9\text{H}_2\text{O}$) with a nanoparticle diameter of 2-10 nm. In sols they form nanoscale aggregates [11]. In the initial colloidal solutions, the concentration of biogenic cobalt-doped ferrihydrite nanoparticles (Feh_Co) was 1.5 g/l.

The reaction of duckweed was recorded by the following parameters: the number of fronds, the number of roots, the length of roots, the wet weight of roots and fronds, the growth rate (table 1).

Table 1. Design of the experiment.

Indicators	Nanoparticle concentration, mg/l	Exposure time			
		3 days	5 days	6 days	7 days
Frond number	0	+	+	-	+
	1.5, 4.5, 15.0	+	+	-	+
Growth rate, days	0	+	+	-	+
	1.5, 4.5, 15.0	+	+	-	+
Root number	0	+	+	-	+
	1.5, 4.5, 15.0	+	+	-	+
Root length, cm	0	+	-	+	-
	1.5, 4.5, 15.0	+	-	+	-
Frond weight, mg	0	-	-	-	+
	1.5, 4.5, 15.0	-	-	-	+
Root weight, mg	0	-	-	-	+
	1.5, 4.5, 15.0	-	-	-	+

Statistical processing of the obtained data was carried out using the package of application programs STATISTICA 10 (StatSoft). In this paper, the data of quantitative parameters of the plants are shown as an arithmetic mean with a standard deviation.

3. Results and discussion

The study of the impact of biogenic cobalt-doped ferrihydrite nanoparticles showed insufficient effect on the increase in the number of fronds in the first days of observation in all concentrations used. However, by the end of the 7-day of exposure was observed a significant decrease in the number of fronds at a maximum concentration of nanoparticles (15.0 mg/l) compared to the control (figure 1).

The growth rate of duckweed was calculated on the basis of data of the fronds number at the initial and final stages of the experiment. The time of doubling the number of duckweed fronds was 0.52 days in the control at a concentration of 1.5 mg/l – 0.50 days; at a concentration of 4.5 mg/l – 0.51 days, it showed slow growth in the frond number growth rate, which manifested at a concentration of 15.0 mg/l nanoparticles, when the doubling time was 0.46 days.

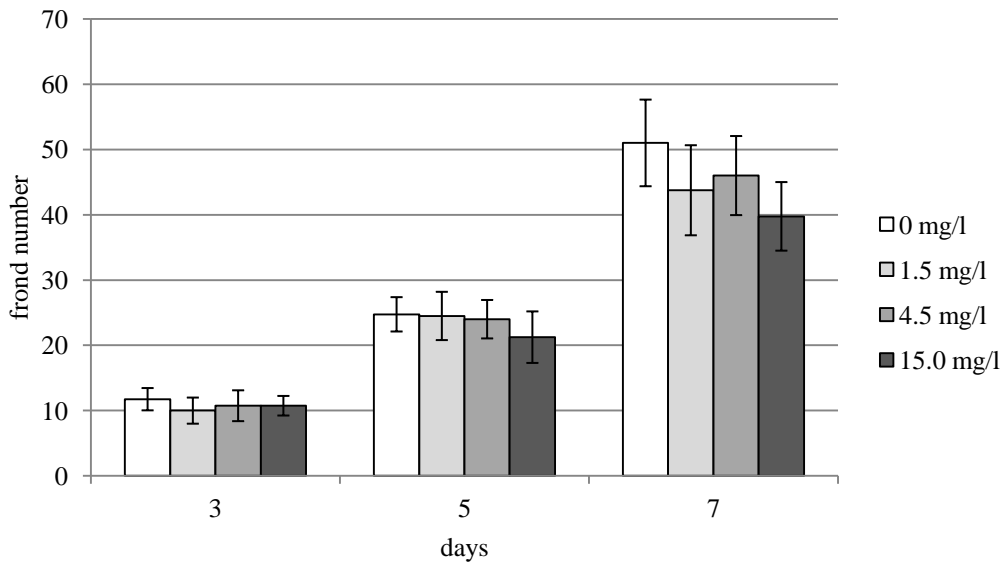


Figure 1. Dynamics of quantities of Lemna minor fronds at different concentrations of biogenic cobalt-doped ferrihydrite nanoparticles.

Analysis of the dynamics of the number of duckweed roots under the action of biogenic cobalt-doped ferrihydrite nanoparticles showed no significant effect compared to control plants (figure 2).

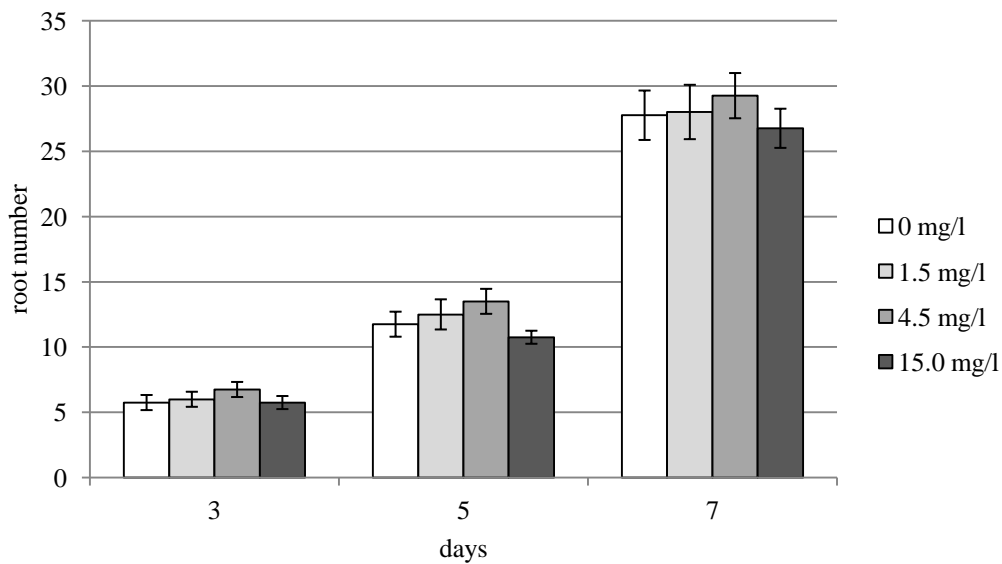


Figure 2. Dynamics of the number of Lemna minor roots at different concentrations of biogenic cobalt-doped ferrihydrite nanoparticles.

Root growth in the presence of biogenic cobalt-doped ferrihydrite nanoparticles was similar to control samples (figure 3).

The results of measuring the weights of the ends and roots under experimental conditions are presented in figure 4. It shows a tendency in decrease of a total biomass of duckweed plants with the increase of nanoparticles concentration in biogenic ferrihydrite, doped with cobalt.

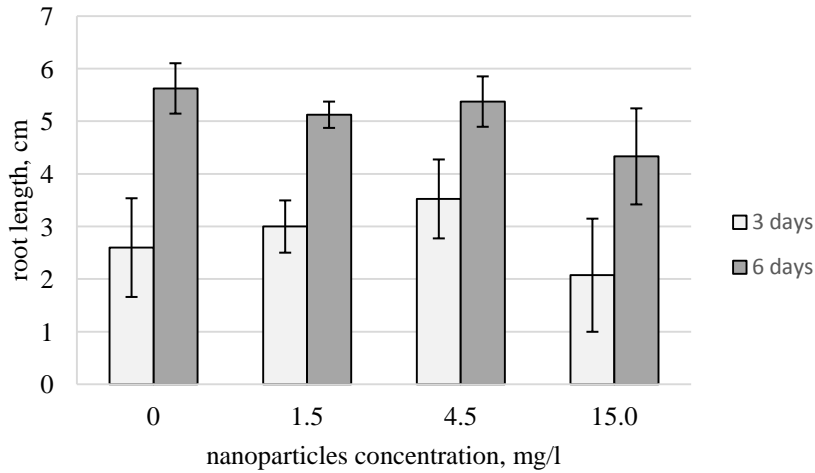


Figure 3. Dynamics of Lemna minor root length growth at different concentrations of biogenic cobalt-doped ferrihydrite nanoparticles.

At the same time, there was no significant difference even at the maximum concentration used in this experiment compared to the control. However, considering the biomass of only fronds, it is possible to note a significant difference with the control at minimum and maximum concentrations. There were no significant differences in the biomass of the roots.

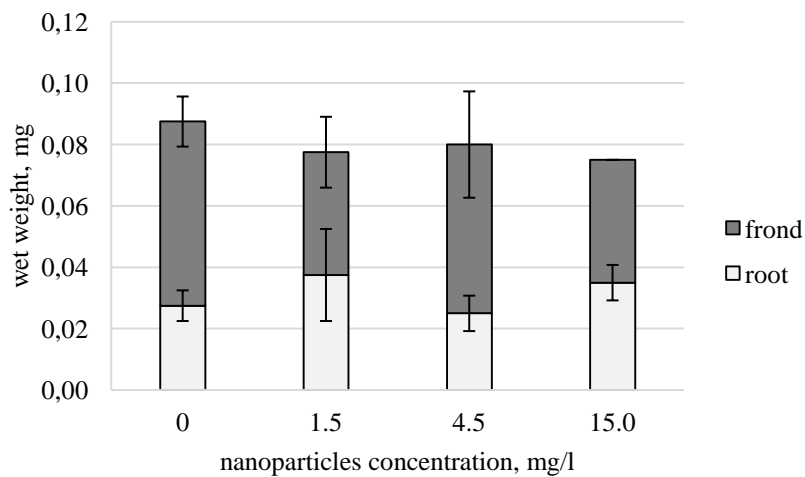


Figure 4. The wet weight of Lemna minor roots and fronds on the 7th day of exposure at different concentrations of biogenic cobalt-doped ferrihydrite nanoparticles.

The study of duckweed growth in a medium with biogenic cobalt-doped ferrihydrite nanoparticles revealed a suppressive effect for a number of indicators (table 2). The effect of nanoparticles manifested at the maximum concentration (15.0 mg/l) reducing the growth of fronds number during 7 days of exposure, which resulted in decreased time for doubling the fronds, as well as decreased biomass of the fronds themselves.

Table 2. The effect of biogenic cobalt-doped ferrihydrite nanoparticles on the growth of Lemna minor.

Indicator	Concentration of nanoparticles, mg/l	Significance level	Effect
Frond number	1.5; 4.5	no	absent
	15.0	<0.05	suppression
Growth rate	1.5; 4.5	no	absent
	15.0	<0.05	suppression
Root number	1.5; 4.5; 15.0	no	absent
Root length	1.5; 4.5; 15.0	no	absent
Frond weight	1.5	<0.05	suppression
	4.5	no	absent
	15.0	<0.01	suppression
Root weight	1.5; 4.5; 15.0	no	absent

4. Conclusion

The results of the experimental study of the reaction of Lemna minor in the presence of biogenic cobalt-doped ferrihydrite nanoparticles medium require further studies due to possible longer-term chronic effect, with the employment of functional activity analysis on the tested organisms.

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